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☐ 1. Document ID: US 20050042735 A1**Using default format because multiple data bases are involved.**

L2: Entry 1 of 84

File: PGPB

Feb 24, 2005

PGPUB-DOCUMENT-NUMBER: 20050042735

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20050042735 A1

TITLE: Metabolic engineering for enhanced production of chitin and chitosan in microorganisms

PUBLICATION-DATE: February 24, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Deng, Ming-De	Manitowoc	WI	US	
McMullin, Thomas W.	Manitowoc	WI	US	
Grund, Alan D.	Manitowoc	WI	US	

US-CL-CURRENT: 435/85; 435/254.2

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC	Draw D
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☐ 2. Document ID: US 20050031584 A1

L2: Entry 2 of 84

File: PGPB

Feb 10, 2005

PGPUB-DOCUMENT-NUMBER: 20050031584

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20050031584 A1

TITLE: Interleukin-2:remodeling and glycoconjugation of interleukin-2

PUBLICATION-DATE: February 10, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
DeFrees, Shawn	North Wales	PA	US	
Zopf, David	Wayne	PA	US	
Bayer, Robert	San Diego	CA	US	
Bowe, Caryn	Doylestown	PA	US	

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L2: Entry 42 of 84

File: USPT

Mar 18, 2003

US-PAT-NO: 6534294

DOCUMENT-IDENTIFIER: US 6534294 B1

**** See image for Certificate of Correction ****

TITLE: Production of chitosan-and chitin-like exopolymers

DATE-ISSUED: March 18, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Lee; Jin Woo	Pusan			KR
Yeomans; Walter G.	Framingham	MA		
Allen; Alfred L.	Pascoag	RI		
Deng; Fang	Drexel Hill	PA		
Gross; Richard A.	Plainview	NY		
Kaplan; David L.	Concord	MA		

US-CL-CURRENT: [435/101](#); [536/102](#), [536/112](#), [536/123](#), [536/126](#), [536/56](#)

CLAIMS:

What is claimed is:

1. A glucose:glucosamine copolymer wherein glucosamine is present at a mole percent in a range of between about 5 and about 65 percent.
2. A method for producing glucose:glucosamine copolymer comprising culturing a biosynthetic agent in medium comprising glucosamine, wherein glucosamine is present in said copolymer at a mole percent in a range of between about 5 and about 65 percent.
3. The method of claim 2, wherein the biosynthetic agent is a microorganism.
4. The method of claim 3 wherein the microorganism is selected from the group consisting of: *Acetobacter xylinum*, *Agrobacter tumefaciens*, *Sarcina ventriculi*, *Rhizobium leguminosarum*, *Mucor rouxii* and *Acinetobacter calcoaceticus*.

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☐ 1. Document ID: US 20040265953 A1

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L13: Entry 1 of 6

File: PGPB

Dec 30, 2004

PGPUB-DOCUMENT-NUMBER: 20040265953

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040265953 A1

TITLE: Production and use of inducible enzymes from trichoderma and bacteria for control of plant pests and for industrial processes

PUBLICATION-DATE: December 30, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Harman, Gary E.	Geneva	NY	US	
Donzelli, Bruno	Geneva	NY	US	
Deng, Shiping	Stillwater	OK	US	

US-CL-CURRENT: [435/69.1](#); [435/183](#), [435/252.3](#)

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KIMC	Draw Ds
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☐ 2. Document ID: US 20040091976 A1

L13: Entry 2 of 6

File: PGPB

May 13, 2004

PGPUB-DOCUMENT-NUMBER: 20040091976

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20040091976 A1

TITLE: Process and materials for production of glucosamine and N-acetylglucosamine

PUBLICATION-DATE: May 13, 2004

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Deng, Ming-De	Manitowoc	WI	US	
Angerer, J. David	Hockessin	DE	US	
Cyron, Don	Lincoln University	PA	US	
Grund, Alan D.	Manitowoc	WI	US	

Jerrell, Thomas A. JR.	Manitowoc	WI	US
Leanna, Candice	Green Bay	WI	US
Mathre, Owen	Wilmington	DE	US
Rosson, Reinhardt	Manitowoc	WI	US
Running, Jeff	Manitowoc	WI	US
Severson, Dave	Two Rivers	WI	US
Song, Linsheng	Manitowoc	WI	US
Wassink, Sarah	Sheboygan	WI	US

US-CL-CURRENT: 435/84; 435/193, 435/252.3

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KIMC	Drawings
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☐ 3. Document ID: US 20020069431 A1

L13: Entry 3 of 6

File: PGPB

Jun 6, 2002

PGPUB-DOCUMENT-NUMBER: 20020069431

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020069431 A1

TITLE: Effect of endochitinase and chitobiosidase and their encoding genes on plant growth and development

PUBLICATION-DATE: June 6, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Broadway, Roxanne M.	Grass Valley	CA	US	
Gongora, Carmenza E.	La Francia		CO	

US-CL-CURRENT: 800/290; 435/200, 800/288

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KIMC	Drawings
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☐ 4. Document ID: US 6838239 B1

L13: Entry 4 of 6

File: USPT

Jan 4, 2005

US-PAT-NO: 6838239

DOCUMENT-IDENTIFIER: US 6838239 B1

TITLE: Chitobiase as a reporter enzyme

DATE-ISSUED: January 4, 2005

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Zyskind, Judith W.	La Jolla	CA		

US-CL-CURRENT: 435/6; 435/183, 435/195, 435/200, 435/206, 435/207, 435/209,
435/252.3, 435/320.1, 435/4, 435/69.1, 536/23.2, 536/23.4, 536/23.7, 536/24.1

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw De
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☐ 5. Document ID: US 6620585 B1

L13: Entry 5 of 6

File: USPT

Sep 16, 2003

US-PAT-NO: 6620585

DOCUMENT-IDENTIFIER: US 6620585 B1

TITLE: Use of ectoenzymes and secreted enzymes to monitor cellular proliferation

DATE-ISSUED: September 16, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Zyskind; Judith W.	La Jolla	CA		

US-CL-CURRENT: 435/6; 435/252.3, 435/252.34, 435/375, 536/24.5

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw De
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☐ 6. Document ID: US 6069299 A

L13: Entry 6 of 6

File: USPT

May 30, 2000

US-PAT-NO: 6069299

DOCUMENT-IDENTIFIER: US 6069299 A

**** See image for Certificate of Correction ****

TITLE: Fungus and insect control with chitinolytic enzymes

DATE-ISSUED: May 30, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Broadway; Roxanne M.	Phelps	NY		
Harman; Gary E.	Geneva	NY		

US-CL-CURRENT: 800/279; 435/200, 435/252.3, 435/320.1, 435/418, 435/419, 435/468,
435/69.1, 536/23.7, 800/288, 800/298, 800/302, 800/306, 800/307, 800/308, 800/309,
800/310, 800/312, 800/313, 800/314, 800/320, 800/321, 800/323

Full	Title	Citation	Front	Review	Classification	Date	Reference			Claims	KWIC	Draw De
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L13: Entry 4 of 6

File: USPT

Jan 4, 2005

DOCUMENT-IDENTIFIER: US 6838239 B1

TITLE: Chitobiase as a reporter enzyme

Detailed Description Text (13):

Chitobiase is one of two enzymes that hydrolyze chitin, an abundant insoluble polysaccharide, to its monomeric unit, N-acetylglucosamine (GlcNac). Chitobiase is known to be present in a number of organisms. For example, the chitobiase enzyme is known to be present in various genera including Arabidopsis, Bacillus, Bombyx, Bos, Caenorhabditis, Candida, Dictyostelium, Entamoeba, Felis, Homo, Korat, Lactobacillus, Leishmania, Mus, Pisum, Porphyromonas, Pseudoalteromonas, Rattus, Serratia, Streptomyces, Sus, Trichoderma, and Vibrio. Specific examples of organisms known to contain chitobiase include Alteromonas sp. 0-7, Arabidopsis thaliana, Bacillus subtilis, Bombyx mori, Bos taurus, Caenorhabditis elegans, Candida albicans, Dictyostelium discoideum, Entamoeba histolytica, Felis catus, Homo sapiens, Korat cats, Lactobacillus casei, Leishmania donovani, Mus musculus, Pisum sativum, Porphyromonas gingivalis, Pseudoalteromonas sp. S9, Rattus norvegicus, Serratia marcescens, Streptomyces plicatus, Streptomyces thermoviolaceus, Sus scrofa, Trichoderma harzianum, Vibrio furnissii, Vibrio harveyi, Vibrio parahaemolyticus, and Vibrio vulnificus.

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L13: Entry 6 of 6

File: USPT

May 30, 2000

DOCUMENT-IDENTIFIER: US 6069299 A

**** See image for Certificate of Correction ****

TITLE: Fungus and insect control with chitinolytic enzymes

Detailed Description Text (2):

Chitin, an insoluble linear .beta.-1,4-linked polymer of N-acetyl-.beta.-D-glucosamine, is a structural polysaccharide that is present in all arthropods, yeast, most fungi, and some stages of nematodes. Chitinolytic enzymes are proteins that catalyze the hydrolysis of chitin by cleaving the bond between the C1 and C4 of two consecutive N-acetylglucosamines. There are three types of chitinolytic enzyme activity: (1) N-acetyl-.beta.-glucosaminidase (i.e., EC 3.2.1.30, abbreviated glucosaminidase), which cleaves monomeric units from the terminal end of chitin, (2) 1,4-.beta.-chitobiosidase (i.e., abbreviated chitobiosidase), which cleaves dimeric units from the terminal end of chitin, and (3) endochitinase (EC 3.2.1.14), which randomly cleaves the chitin molecule internally (Sahai, et al., "Chitinases of Fungi and Plants: Their Involvement in Morphogenesis and Host-Parasite Interaction," FEMS Microbiol. Rev., 11:317-38 (1993), which is hereby incorporated by reference). Two or three types of enzymes are often synthesized by a single organism (Harman, et al., "Chitinolytic Enzymes of *Trichoderma harzianum*: Purification of Chitobiase and Endochitinase," Phytopathology, 83:313-18 (1993), Neugebauer, et al., "Chitinolytic Properties of *Streptomyces lividans*," Arch. Microbiol., 156:192-97 (1991), Romaguera, et al., "Protoplast Formation by a Mycolase from *Streptomyces olivaceoviridis* and Purification of Chitinases," Enzyme Microb. Technol., 15:412-17 (1993), which are hereby incorporated by reference), which may enhance the speed and/or efficiency of degradation of chitin.

Detailed Description Text (77):

In general, secretion of fungal and bacterial chitinolytic enzymes is regulated by availability of carbon (e.g., chitodextrins); the presence of glucose or N-acetylglucosamine represses secretion of chitinolytic enzymes, while chitin induces the secretion of these enzymes (Harman, et al., "Chitinolytic Enzymes of *Trichoderma harzianum*: Purification of Chitobiase and Endochitinase," Phytopathology, 83:313-18 (1993), Monreal, et al., "The Chitinase of *Serratia Marcescens*," Can. J. Microbiol., 15:689-96 (1969), St. Leger, et al., "Cuticle-Degrading Enzymes of Entomopathogenic Fungi: Regulation of Production of Chitinolytic Enzymes," J. Gen. Microbiol., 132:1509-17 (1986), Ulhoa, et al., "Regulation of Chitinase Synthesis in *Trichoderma harzianum*," J. Gen. Microbiol., 137:2163-69 (1991), which are hereby incorporated by reference). In support of these previous reports, the presence of chitin in the liquid medium resulted in a minimum of a 5-fold increase of all three types of enzyme activity secreted by *S. albidoflavus* (Table 2).

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<input type="checkbox"/>	L11	trichoderma same acetylglucosamine	7
<input type="checkbox"/>	L10	Fungus same bacteria same chitin.clm.	1
<input type="checkbox"/>	L9	Fungus same bacteria same chitin	179
<input type="checkbox"/>	L8	L6 and L7	0
<input type="checkbox"/>	L7	n-acetylhexosaminidase	57
<input type="checkbox"/>	L6	beta 1 3 glucanase	103
<input type="checkbox"/>	L5	n-acetylhexosaminidase same chitin.clm.	0
<input type="checkbox"/>	L4	n-acetylhexosaminidase.clm.	3
<input type="checkbox"/>	L3	n-acetylhexosamidase.clm.	0
<input type="checkbox"/>	L2	L1 and bacteria	84
<input type="checkbox"/>	L1	N-acetylglucosamine same fungus	126

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L2 1 N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGI

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L2 ANSWER 1 OF 1 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 2004-09830 BIOTECHDS

TITLE: Producing glucosamine or N-acetylglucosamine by fermentation involves culturing microorganism comprising glucosamine-6-phosphate acetyltransferase, in fermentation medium, and collecting product;
glucosamine and N-acetylglucosamine production via recombinant bacterium and fungus culture

AUTHOR: DENG M; ANGERER J D; CYRON D; GRUND A D; JERRELL T A; LEANNA C; MATHRE O; ROSSON R; RUNNING J; SEVERSON D; SONG L; WASSINK S

PATENT ASSIGNEE: ARKION LIFE SCI LLC

PATENT INFO: WO 2004003175 8 Jan 2004

APPLICATION INFO: WO 2003-US20925 1 Jul 2003

PRIORITY INFO: US 2002-393348 1 Jul 2002; US 2002-393348 1 Jul 2002

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2004-203380 [19]

AB DERWENT ABSTRACT:

NOVELTY - Producing (M1) glucosamine or N-acetylglucosamine by fermentation comprising culturing in a fermentation medium a microorganism (I) which comprises endogenous glucosamine-6-phosphate acetyltransferase (II) and a genetic modification that increases the activity of (II), and collecting a product produced from the step of culturing, which is chosen from the group consisting of glucosamine-6-phosphate, glucosamine, is new.

DETAILED DESCRIPTION - Producing (M1) glucosamine or N-acetylglucosamine by fermentation involves (a) culturing in a fermentation medium a microorganism (I) which comprises endogenous glucosamine-6-phosphate acetyltransferase (II) and a genetic modification that increases the activity of (II), glucosamine-6-phosphate synthase (III) or glucosamine-6-phosphate deaminase (IV), or decreases the activity of (IV) and increases the activity of glucosamine-1 phosphate N-acetyltransferase (V), and (b) collecting a product produced from the step of culturing, which is chosen from the group consisting of glucosamine-6-phosphate, glucosamine, glucosamine-1-phosphate, N-acetylglucosamine-1-phosphate, N-acetylglucosamine-6-phosphate, and N-acetylglucosamine. INDEPENDENT CLAIMS are also included for the following: (1) a genetically modified microorganism (VI) comprising a genetic modification that increases the activity of (II); (2) a genetically modified microorganisms (VII) comprising a genetic modification that increases the activity of (IV); (3) a genetically modified microorganisms (VIII) comprising a genetic modification that decreases the activity of (IV) and increases the activity of (V); (4) producing (M2), N-acetylglucosamine involves obtaining a fermentation broth containing solubilized N-acetylglucosamine that is a product of a fermentation process, and recovering N-acetylglucosamine-containing solids from the fermentation broth; (5) producing (M3), glucosamine form a source of N-acetylglucosamine, comprising obtaining a source of N-acetylglucosamine from N-acetylglucosamine, N-acetylglucosamine-6-phosphate or N-acetylglucosamine-1-phosphate and treating the source of N-acetylglucosamine to produce a glucosamine product such as glucosamine, glucosamine-6-phosphate and glucosamine-1-phosphate; and (6) producing (M4), glucosamine by fermentation comprising culturing (I) in a fermentation medium, and collecting a product such as glucosamine-6-phosphate or glucosamine which is produced by the culturing step, where (I) has been transformed with a recombinant nucleic acid molecule comprising a nucleic acid sequence encoding glucosamine-6-phosphate, the expression of the recombinant nucleic acid molecule is controlled by a lactose induction, and the culturing step involves growing the microorganism in the fermentation medium comprising glucose

as a carbon source at a pH 4.5-7 and at a temperature of 25-37 degrees Centigrade, inducing transcription of the nucleic acid sequence by addition of lactose to the fermentation medium in the absence of adding additional glucose to the medium, and fermenting the microorganism after the inducing step, in the presence of lactose at pH 4.5-6.7 and at temperature 25-37 degrees Centigrade.

BIOTECHNOLOGY - Preferred Method: (M1) further involves recovering a product, dephosphorylating the product such as glucosamine-6-phosphate and glucosamine-1-phosphate to produce glucosamine, or dephosphorylating the product such as N-glucosamine-6-phosphate and N-glucosamine-1-phosphate to produce N-acetylglucosamine, and treating a product such as N-acetylglucosamine, N-acetylglucosamine-6-phosphate and N-acetylglucosamine-1-phosphate, under acid and heat conditions or by enzymatic deacetylation to produce a glucosamine product such as glucosamine, glucosamine-6-phosphate or glucosamine-1-phosphate. N-glucosamine produced by the fermentation is recovered by precipitating or crystallizing N-glucosamine-containing solids from the fermentation broth. In (M1), the culturing step includes the step of maintaining the carbon source at a concentration of 0.5%-5% in the fermentation medium. The fermentation medium comprises yeast extract, glucose and ribose or gluconic acid. The carbon source in the fermentation medium is chosen from glucose, fructose, a pentose sugar, lactose and gluconic acid, where the pentose sugar is ribose, xylose or arabinose. The culturing step is performed at a temperature of 25-45 degrees Centigrade, preferably 37 degrees Centigrade, and at a pH of 4-7.5, preferably 6.7-7.5 or 4.5-5. The microorganism is chosen from **bacteria** and **fungi**, or **bacteria** and yeast. The bacterium is chosen from a genus *Escherichia*, *Bacillus*, *Lactobacillus*, *Pseudomonas* and *Streptomyces*. The **fungi** is chosen from a genus *Aspergillus*, *Absidia*, *Rhizopus*, *Chrysosporium*, *Neurospora* and *Trichoderma*. The yeast is chosen from genus *Saccharomyces*, *Candida*, *Hansenula*, *Pichia*, *Klveromyces* and *Phaffia*. In (M1), the collecting step involves recovering an intracellular product from the microorganism chosen from intracellular glucosamine-6-phosphate, glucosamine-1-phosphate, N-acetylglucosamine-6-phosphate, N-acetylglucosamine-1-phosphate, N-acetylglucosamine and glucosamine or recovering an extracellular product from the fermentation medium chosen from glucosamine and N-acetylglucosamine. In (I) of (M1), the genetic modification to increase the activity of (II), (V) or glucosamine-6-phosphate N-acetyltransferase (IX) provides a result chosen from increased enzymatic activity of (II), (V) or (IV), overexpression of (II), (V) or (IV) by the microorganisms, reduced N-acetylglucosamine-6-phosphate product inhibition of (II), (V) or (IV) and increased affinity of (II), (V) or (IV) for glucosamine-6-phosphate. (I) is transformed with a recombinant nucleic acid molecule comprising a nucleic acid sequence encoding (II), where the nucleic acid encoding (II) has a genetic modification which increases the enzymatic activity of (II). (II) has enzyme activity and comprises a fully defined sequences (S1) of 159, 149 or 149 amino acids as given in the specification, or an amino acid sequence that is at least 35%, 50% or 70% identical to (S1). The expression of the recombinant nucleic acid molecule is inducible by lactose. (I) further comprises a genetic modification to reduce inhibition of transcription induction by lactose, where the genetic modification comprises a partial or complete deletion or inactivation of a gene encoding a LacI repressor protein. (I) further comprises a genetic modification that increases the activity of (III), phosphoglucoisomerase in microorganisms, a glutamine synthetase or glucose-6-phosphate dehydrogenase, or decreases the activity of (IV). (III) has a modification to reduce product inhibition of (III) as compared to the wild-type glucosamine-6-phosphate synthase. (III) has enzymatic activity and comprises a sequences (S2) chosen from 6 fully defined sequence of 609 amino acids or a fully defined sequence of 600, 717 or 713 amino acids as given in the specification, or an amino acid sequence that is at least 35%, 50% or 70% identical to (S2). (III) preferably contains a sequence chosen from 6 fully defined sequence of 609 amino acids as given in the specification. (I) is transformed with a recombinant nucleic acid molecule comprising a nucleic acid sequence encoding (III), or phosphoglucoisomerase, glutamine synthetase or glucose-6-phosphate dehydragenase which comprises a fully defined sequence of 549, 469 or 491 amino acids respectively, as given in the specification. The genetic

modification to decrease or increase the activity of (IV) involves a partial or complete deletion or inactivation of an endogenous gene encoding (IV) in the micro organism. (I) further comprises a partial or complete deletion, or inactivation of phosphofructokinase or genes encoding enzymes responsible for glycogen synthesis in the microorganisms, where the genes encoding enzymes responsible for glycogen synthesis comprise ADP-glucose pyrophosphorylase, glycogen synthase and a branching enzyme. The genetic modifications do not inhibit the ability of (I) to metabolize galactose. In (I), the genetic modification that increases the activity of (IV), provides a results chosen from overexpression of (IV), by the microorganisms, increased enzymatic activity of (IV), increased reverse reaction or reduced forward reaction of (IV) to form glucosamine-6-phosphate, increased or reduced affinity of (IV), for glucosamine-6-phosphate, and reduced glucosamine-6-phosphate product inhibition of (IV). (I) is transformed with a recombinant nucleic acid molecule comprising a nucleic acid sequences encoding (IV) which has a genetic modification to increase the enzymatic activity of (IV), nucleic acid sequence encoding glucosamine-1-phosphate N-acetyltransferase/N-acetylglucosamine-1-phosphate uridyltransferase (X), nucleic acid sequence encoding truncated (X) having, glucosamine-1-phosphate N-acetyltransferase (XI) activity and reduced or no N-acetylglucosamine-1-phosphate uridyltransferase activity, or nucleic acid sequence encoding (XI), where (X) or (XI) has a genetic modification which increases the activity of (X) or (XII). The (IV), (X) or truncated (X) comprises a fully defined sequence of 266 amino acids (S4), 456 amino acids (S4) or 380 amino acids (S5), respectively as given in the specification, or amino acids sequence that is at least 35% identical to (S3), (S4) or (S5) respectively. (I) further comprises a genetic modification to decrease the activity of (III) and/or increase the activity of (VI), (V) or bifunctional (X). The (IX) comprises (S1) or amino acids sequence identical to (S1). (M2) further involves removing the cellular material from the fermentation broth, contacting the fermentation broth with an ion exchange resin such as anion and a cation exchange resin or mixed bed of anion and cation exchange resins, decolorizing the fermentation broth which is chosen from multiple N-acetylglucosamine crystallizations, activated carbon treatment and chromatographic decolorization, and recovering the product by precipitating or crystallizing N-acetylglucosamine-containing solids from the fermentation broth, and concentrating the fermentation broth containing solubilized N-acetylglucosamine. The concentration step is conducted at less than atmospheric pressure by membrane separation, at a temperature of 40-75 degrees Centigrade, preferably 45-55 degrees Centigrade. The concentrating step is conducted to achieve a solids content in the fermentation broth of at least 40% or 45% solids. (M2) further involves cooling the fermentation broth after concentrating step, at -5 degrees Centigrade-45 degrees Centigrade, preferably between -5 degrees Centigrade and room temperature, most preferably room temperature. (M2) further involves seeding the fermentation broth with crystals of N-acetylglucosamine, recovering the product by contacting N-acetylglucosamine with a water miscible solvent, drying the recovered N-acetylglucosamine containing solids and washing it with a water miscible solvent. (M2) further involves dissolving the recovered N-acetylglucosamine-containing solids to form an N-acetylglucosamine solution and recovering N-acetylglucosamine-containing solids from the solution, and filtering the fermentation broth to remove bacterial endotoxins. The seed crystals are chosen from the group consisting of N-acetylglucosamine crystals formed by nucleation in the fermentation broth and externally provided N-acetylglucosamine crystals. The water miscible solvent is isopropyl alcohol (IPA) ethanol, methanol, acetone, tetrahydrofuran, dimethylsulfoxide, dimethylformamide, dioxane and acetonitrile. In (M3), the source of N-acetylglucosamine is at least 40% N-acetylglucosamine of dry solids in the source, and is produced by a fermentation process. The source of N-acetylglucosamine is provided as a solid or in suspended in a solution such as aqueous, low-boiling, primary or secondary alcohol. The treating step involves contacting the source of N-acetylglucosamine with a deacetylating enzyme in the presence of aqueous sodium or calcium chloride solution, or alcohol to esterify the alcohol, to produce the glucosamine product, and hydrolyzing the source of N-acetylglucosamine under acid using hydrochloric acid at a

concentration of 10-40% by weight, and heat conditions, at 60-100 degrees Centigrade, preferably 70-90 degrees Centigrade, the hydrolyzing step is performed for 10 minutes to 24 hours. The ratio of the weight of hydrochloric acid solution to the source of N-acetylglucosamine as a pure dry weight is from 1:1-5:1. The deacetylating enzyme is immobilized on a substrate, and is N-acetylglucosamine-6-P deacetylase or N-acetylglucosamine deacetylase, where the deacetylating enzyme is a **chitin** deacetylase that has been modified to or selected for its ability to deacetylate an N-acetylglucosamine monomer to produce glucosamine. (M3) further involves cooling the hydrolyzed solution at -5 degrees Centigrade-40 degrees Centigrade to precipitate the glucosamine hydrochloride and recovering the precipitated glucosamine hydrochloride-containing solids from the solution, where the recovering step involves collecting the precipitated glucosamine hydrochloride-containing solids, washing it with water miscible solvent, drying the glucosamine hydrochloride-containing solids, dissolving the solids in water to form a solution, adjusting the pH of the solution to 2.5-4, contacting the solution with activated carbon to decolorize the glucosamine hydrochloride-containing solids, removing the activated carbon from the solution and crystallizing glucosamine hydrochloride from the solution which involves concentrating the glucosamine hydrochloride at a temperature of less than 50 degrees Centigrade, less than atmospheric pressure. The hydrolyzing step is performed by continuously blending the source of N-acetylglucosamine with a hydrochloric acid solution or a recycled hydrolysis mother liquor to maintain the source of N-acetylglucosamine as a dissolved solution, followed by addition of anhydrous hydrochloric acid under heat conditions to initiate hydrolysis and convert the **N-acetylglucosamine** to glucosamine hydrochloride. The hydrolysis step is performed at temperature 60-100degrees and at the solution boiling point at one atmosphere. The recovery step involves adding primary or secondary alcohol such as methanol, isopropanol, ethanol, n-propanol, n-butanol or sec-butanol, to the hydrolysis solution before the hydrolysis step. The drying of crystallized glucosamine hydrochloride is conducted at less than 70 degrees Centigrade for less than 6 hours with an air sweep. (M3) involves a further step of removing the acetic acid ester formed with the alcohol following the hydrolysis step, before recycling the hydrolysis solution for reuse, where the acetic acid ester is removed by distillation, flashing or concentration at less than atmospheric pressure. (M3) further involves crystallization, precipitation, mixing a salt with the glucosamine product and contacting the obtain mixture with an ion exchange medium. The salt is chloride, phosphate, a sulfate, an iodide or a bisulfate. In (M4), trace elements include iron and the organisms are grown at pH 6.1. The organisms are fermented in the presence of glucose at a pH 4.5-5, preferably 6.7. Preferred Microorganisms: (VI) further comprises genetic modification that increase the activity of (III), or decrease the activity of (IV). (VII) further comprises genetic modification that increase the activity of (IX) or (V), or decrease the activity of (III). (VIII) further comprises a genetic modification that increase activity of (III).

USE - (M1) is useful for producing glucosamine or N-acetylglucosamine by fermentation (claimed).

EXAMPLE - Recombinant Escherichia coli was inoculated into the fermentation medium comprising 5 to 10 g/l-1 of lactose, and 65% of glucose which was fed into the fermentation medium at 6.5 gl-1 hr-1, at pH 6.9 and a temperature of 37 degrees Centigrade. The 20% of oxygen was controlled by agitation. The fermentation was allowed for 60-72 hours and the fermentation broth was obtained which was subjected to filtration and micro-filtration to remove the cells. Depending on the fermentation conditions, the percentage of N-acetylglucosamine in the dissolved solid ranged from 70 to 87%. The crude N-acetylglucosamine product was subjected to a combination of cation and anion deionization steps to increase N-acetylglucosamine purity in the crude broth. Activated carbon was added to a fermentation sample (30 g/l) containing 87% (w) N-acetylglucosamine in the dissolved solid and the mixture was stirred at room temperature for an hour followed by filtration using medium filter paper. The filtrate showed reduced color and was pale yellowish brown. The amount of solid was measured and the percentage of N-acetylglucosamine in the solid was 88%. The carbon treated sample

containing 80.5 g solid was concentrated at 45 degrees Centigrade-50 degrees Centigrade vacuum to 45% (w) solid and shaken at room temperature for 16 hours. The precipitate was collected by filtration on medium filter paper and washed with ethanol. After drying under vacuum, 33.2 grams of white solid was obtained. The obtained white solid was 100% pure N-acetylglucosamine and the recovery was 47%. (326 pages)

=> s chitin and bacteria and fungi

L3 708 CHITIN AND BACTERIA AND FUNGI

=> dup rem l3

PROCESSING COMPLETED FOR L3

L4 531 DUP REM L3 (177 DUPLICATES REMOVED)

=> s l4 and chitinolytic

L5 69 L4 AND CHITINOLYTIC

=> s l5 and (acetylhexosaminidase or beta-1 3 glucanase)

L6 10 L5 AND (ACETYLHEXOSAMINIDASE OR BETA-1 3 GLUCANASE)

=> d l6 1-10 ibib ab

L6 ANSWER 1 OF 10 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:459077 HCAPLUS

DOCUMENT NUMBER: 139:273320

TITLE: The **chitinolytic** activity of *Bacillus Cohn* **bacteria** antagonistic to phytopathogenic **fungi**

AUTHOR(S): Aktuganov, G. E.; Melent'ev, A. I.; Kuz'mina, L. Yu.; Galimzyanova, N. F.; Shirokov, A. V.

CORPORATE SOURCE: Inst. Biology, Ufa Res. Center, Russian Academy Sciences, Ufa, 450054, Russia

SOURCE: Microbiology (Moscow, Russian Federation) (Translation of *Mikrobiologiya*) (2003), 72(3), 313-317
CODEN: MIBLAO; ISSN: 0026-2617

PUBLISHER: MAIK Nauka/Interperiodica Publishing

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Among the 70 tested *Bacillus* spp. strains antagonistic to phytopathogenic **fungi**, 19 were found to possess **chitinolytic** activity when grown on solid media with 0.5% colloidal **chitin**. The **chitinolytic** activity of almost all of these 19 strains grown in liq. cultures ranged from 0.1 to 0.3 U/mL. One of the 19 strains exhibited exochitinase activity. In addn. to chitinase, two strains also produced chitosanase and one strain, **beta**-1, **3-glucanase**. No correlation was found between the antifungal activity of the bacillar strains studied and their ability to synthesize extracellular chitinase. Among the 19 **chitinolytic** strains, the correlation between these parameters was also low (rx, y = 0.45), although the enzymic preps. of most of these strains inhibited the growth of the phytopathogenic fungus *Helminthosporium sativum*.

REFERENCE COUNT: 15 THERE ARE 15 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 2 OF 10 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1999:708877 HCAPLUS

DOCUMENT NUMBER: 131:334542

TITLE: *Lytobacter mycophilus*, a novel **chitinolytic** bacterium that inhibits the growth of phytopathogenic **fungi**

INVENTOR(S): Kobayashi, Donald Y.; Holtman, Michael A.

PATENT ASSIGNEE(S): Rutgers, the State University of New Jersey, USA

SOURCE: PCT Int. Appl., 117 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9955833	A2	19991104	WO 1999-US9032	19990427
WO 9955833	A3	19991216		
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG AU 9936660 A1 19991116 AU 1999-36660 19990427 PRIORITY APPLN. INFO.: US 1998-83119P P 19980427 WO 1999-US9032 W 19990427				

AB A novel bacterial genus, *Lytobacter*, and its representative species, *Lytobacter mycophilus*, are disclosed. *Lytobacter mycophilus* strains are isolated from soil and display antifungal characteristics, specifically against *Magnaporthe poae*, due to the prodn. of several lytic enzyme activities, including chitinase, **.beta.-1,3-glucanase**, lipase/esterase and protease. Sequence comparison 16S rRNAs identifies *Lytobacter* as a member of the *Xanthomonas* group of **.gamma. proteobacteria**. Chitinase and **.beta.-1,3-glucanase** enzymes produced by the novel bacterium are also disclosed. **Bacteria** from this novel genus and isolated antifungal components thereof are expected to be particularly useful for biol. control of various fungal diseases of plants, particularly summer patch disease of turfgrass. The bacterium was isolated during a screen of soil for **chitinolytic bacteria** using sterilized mycelium of *M. poae* as **chitin** source. Testing isolated **bacteria** for their ability to suppress *M. poae* infection of plants identified one species of *Bacillus*, three species of *Serratia*, a *Klebsiella*, and a hitherto unidentified species. The *chiA* chitinase gene of *L. mycophilus* was cloned by probing a *Sau3A* partial digest library in a **.lambda. vector** with a chitinase gene fragment from *Stenotrophomonas maltophilia*. Similarly, a gene for a **.beta.-1,3-glucanase** was cloned using a *Citrobacter* host.

L6 ANSWER 3 OF 10 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1998:77470 HCAPLUS

DOCUMENT NUMBER: 128:153517

TITLE: Antifungal properties of **chitinolytic dune soil bacteria**

AUTHOR(S): De Boer, Wietse; Klein Gunnewiek, Paulien J. A.; Lafeber, Petra; Janse, Jaap D.; Spit, Bendien E.; Woldendorp, Jan W.

CORPORATE SOURCE: Netherlands Institute of Ecology, Centre for Terrestrial Ecology, Department of Plant-Microorganism Interactions, Heteren, 6666 ZG, Neth.

SOURCE: Soil Biology & Biochemistry (1998), 30(2), 193-203
CODEN: SBIOAH; ISSN: 0038-0717

PUBLISHER: Elsevier Science Ltd.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The main objective was a better understanding of the relationship between **chitinolytic** and antifungal properties of **bacteria** that occur naturally in soils, i.e. without artificial selection. Three inner dune sites, two of which were lime-poor and one lime-rich, along the Dutch coast, were selected for this study. **Bacteria** that were able to degrade colloidal **chitin** in water-agar comprised 0.2-5.7% of the total amt. of culturable **bacteria** of these dune sites. *Pseudomonas* were the most abundant culturable, **chitin**-degrading **bacteria** at the lime-poor sites, whereas *Xanthomonas* and *Cytophaga* were important at the lime-rich site. **Chitinolytic** actinomycetes were relatively abundant at all 3 sites. **Chitinolytic** and non-**chitinolytic bacteria** were randomly selected and tested for antagonistic activities against fungal dune strains [*Chaetomium globosum*, *Fusarium culmorum*, *F. oxysporum*,

Idriella (Microdochium), bolleyi, Mucor hiemalis, Phoma exigua, Ulocladium]. The tests were done using water-agar to simulate the energy-limiting conditions that **bacteria** will encounter in dune soils. The percentage of bacterial isolates that were antagonistic against these **fungi** was higher for **chitinolytic** strains than for non-**chitinolytic** ones. Therefore, the possible involvement of chitinase with respect to the inhibition of fungal growth was studied. In many cases, the inhibition of fungal growth was not accompanied by bacterial chitinase prodn. There was also no relationship between the activity of cell wall degrading enzymes (**.beta.-1,3-glucanase** and protease) and antagonism.

Chitinolytic bacteria had selective rather than general antifungal properties, which were not necessarily related to differences in susceptibility of the **fungi** towards antagonism. Antibiotics were involved in the antagonistic activities of **chitinolytic bacteria** against **fungi**. Only growing **fungi** were antagonized by the **chitinolytic bacteria**. None of the **chitinolytic bacteria** were able to lyse existing mycelium of any of the **fungi**. The relevance of the results for the ecol. of **chitinolytic soil bacteria** is discussed.

REFERENCE COUNT: 40 THERE ARE 40 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 4 OF 10 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1998:51693 HCAPLUS

DOCUMENT NUMBER: 128:150915

TITLE: Synergistic, antifungal interactions of **chitinolytic** enzymes from **fungi**, **bacteria** and plants

AUTHOR(S): Lorito, Matteo; Woo, Sheridan Lois; Donzelli, Bruno; Scala, Felice

CORPORATE SOURCE: Ist. di Patologia Vegetale, e Centro di Studio CNR sulle Tecniche di Lotta Biol. (CETELOBI)-Patologia Vegetale, Univ. degli Studi di Napoli "Federico II", Naples, 80055, Italy

SOURCE: Chitin Enzymology, Proceedings of the International Symposium on Chitin Enzymology, 2nd, Senigallia, Italy, May 8-11, 1996 (1996), 157-164. Editor(s): Muzzarelli, Riccardo A. A. Atec Edizioni: Grottammare, Italy.

CODEN: 65LZA3

DOCUMENT TYPE: Conference

LANGUAGE: English

AB **Chitinolytic** enzymes with different modes of action may have synergistic activity when used in combinations. N-acetyl-**.beta.-glucosaminidases** or **chitin 1,4-.beta.-chitobiosidases** from **fungi** or **bacteria** were synergistic with endochitinases from **fungi** or plants in the inhibition of spore germination and hyphal elongation of Botrytis cinerea and other phytopathogenic **fungi**. **Chitinolytic** enzymes were also synergistic with other cell wall degrading enzymes (CWDEs) such as **.beta.-1,3** and **.beta.-1,6 glucanases**. In addn., **chitinolytic** enzymes from different sources may synergistically enhance the inhibitory activity of membrane-affecting compds. (MAC). The mechanism of this interaction was investigated by monitoring the **.beta.-glucan** and the **chitin** synthase activities assocd. with cell membranes. Some MACs altered structure and function of the cell membrane, inhibited the synthesis of major cell wall components in three different fungal systems (Trichoderma, Botrytis and Saccharomyces) and made the cell wall more sensitive to the action of **chitinolytic** enzymes. When MACs and enzymes were applied together against **fungi**, MACs could reduce the ability of the cell to repair the wall and amplify the effect of the enzymes. On the other hand, the enzyme activity, by producing a partial digestion of the cell wall, could facilitate the penetration of the MACs to reach their target at higher concns. The ability to interact with other biol. active compds. indicates that **chitinolytic** enzymes and genes encoding for them may be used as antimicrobial factors against **chitin** -contg. pathogenic **fungi**.

REFERENCE COUNT: 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS

L6 ANSWER 5 OF 10 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
 ACCESSION NUMBER: 2004:173120 BIOSIS
 DOCUMENT NUMBER: PREV200400174292
 TITLE: Mycolytic effect of extracellular enzymes of antagonistic microbes to *Colletotrichum falcatum*, red rot pathogen of sugarcane.
 AUTHOR(S): Viswanathan, R. [Reprint Author]; Sundar, A. Ramesh; Premkumari, S. Merina
 CORPORATE SOURCE: Plant Pathology Section, Sugarcane Breeding Institute, Indian Council of Agricultural Research, Coimbatore, 641007, India
 rasaviswanathan@yahoo.co.in
 SOURCE: World Journal of Microbiology & Biotechnology, (December 2003) Vol. 19, No. 9, pp. 953-959. print.
 ISSN: 0959-3993 (ISSN print).
 DOCUMENT TYPE: Article
 LANGUAGE: English
 ENTRY DATE: Entered STN: 31 Mar 2004
 Last Updated on STN: 31 Mar 2004

AB Strains of selected **bacteria** and *Trichoderma harzianum* isolated from sugarcane rhizosphere and endosphere regions were tested for the production of **chitinolytic** enzymes and their involvement in the suppression of *Colletotrichum falcatum*, red rot pathogen of sugarcane. Among several strains tested for **chitinolytic** activity, 12 strains showed a clearing zone on **chitin**-amended agar medium. Among these, bacterial strains AFG2, AFG 4, AFG 10, FP7 and VPT4 and all the tested *T. harzianum* strains produced clearing zones of a size larger than 10 mm. The antifungal activity of these strains increased when **chitin** was incorporated into the medium. *Trichoderma harzianum* strain T5 showed increased levels of activity of N-acetylglucosaminidase and **beta-1,3-glucanase** when grown on minimal medium containing **chitin** or cell wall of the pathogen. Lytic enzymes of bacterial strains AFG2, AFG4, VPT4 and FP7 and *T. harzianum* T5 inhibited conidial germination and mycelial growth of the pathogen. Enzymes from *T. harzianum* T5 were found to be the most effective in inhibiting the fungus. When mycelial discs of the pathogen were treated with the enzymes, electrolytes were released from fungal mycelia. The results indicated that antagonistic *T. harzianum* T5 caused a higher level of lysis of the pathogen mycelium, and the inhibitory effect was more pronounced when the lytic enzymes were produced using **chitin** or cell wall of the pathogen as carbon source.

L6 ANSWER 6 OF 10 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
 ACCESSION NUMBER: 2003:387650 BIOSIS
 DOCUMENT NUMBER: PREV200300387650
 TITLE: The **chitinolytic** activity of *Bacillus cohn* **bacteria** antagonistic to phytopathogenic **fungi**.
 AUTHOR(S): Aktuganov, G. E. [Reprint Author]; Melent'ev, A. I. [Reprint Author]; Kuz'mina, L. Yu. [Reprint Author]; Galimzyanova, N. F. [Reprint Author]; Shirokov, A. V. [Reprint Author]
 CORPORATE SOURCE: Ufa Research Center, Institute of Biology, Russian Academy of Sciences, Pr. Oktyabrya 69, Ufa, 450054, Russia
 gleakt@anrb.ru
 SOURCE: Mikrobiologiya, (May-June 2003) Vol. 72, No. 3, pp. 356-360. print.
 CODEN: MIKBA5. ISSN: 0026-3656.
 DOCUMENT TYPE: Article
 LANGUAGE: Russian
 ENTRY DATE: Entered STN: 20 Aug 2003
 Last Updated on STN: 20 Aug 2003

AB Among the 70 tested *Bacillus* spp. strains antagonistic to phytopathogenic **fungi**, 19 were found to possess **chitinolytic** activity when grown on solid media with 0.5% colloidal **chitin**. The **chitinolytic** activity of almost all of these 19 strains grown in liquid cultures ranged from 0.1 to 0.3 U/ml. One of the 19 strains

exhibited exochitinase activity. In addition to chitinase, two strains also produced chitosanase and one strain, **beta-1, 3-glucanase**. No correlation was found between the antifungal activity of the bacillar strains studied and their ability to synthesize extracellular chitinase. Among the 19 **chitinolytic** strains, the correlation between these parameters was also low (rx,y=0.45), although the enzymatic preparations of most of these strains inhibited the growth of the phytopathogenic fungus *Helminthosporium sativum*.

L6 ANSWER 7 OF 10 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 2001:8311 BIOSIS
DOCUMENT NUMBER: PREV200100008311
TITLE: Biological control of *Sclerotinia minor* using a **chitinolytic** bacterium and actinomycetes.
AUTHOR(S): El-Tarabily, K. A.; Soliman, M. H.; Nassar, A. H.; Al-Hassani, H. A.; Sivasithamparam, K.; McKenna, F.; St. J. Hardy, G. E. [Reprint author]
CORPORATE SOURCE: School of Biological Sciences and Biotechnology, Murdoch University, Murdoch, W.A., 6150, Australia
g-hardy@central.murdoch.edu.au
SOURCE: Plant Pathology (Oxford), (October, 2000) Vol. 49, No. 5, pp. 573-583. print.
CODEN: PLPAAD. ISSN: 0032-0862.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 21 Dec 2000
Last Updated on STN: 21 Dec 2000

AB Isolates of 85 **bacteria** and 94 streptomycete and 35 nonstreptomycete actinomycetes were obtained from a lettuce-growing field in Al-Ain, United Arab Emirates, on colloidal **chitin** agar, and screened for their ability to produce chitinase. Twenty-three **bacteria** and 38 streptomycete and 15 nonstreptomycete actinomycete isolates produced high levels of chitinase and were examined in vitro for their ability to suppress the growth of *Sclerotinia minor*, a pathogen causing basal drop disease of lettuce. The three most suppressive isolates were examined further for their production of **beta-1,3-glucanase** and antifungal activity as well as their ability to colonize the roots and rhizosphere of lettuce in vitro and in planta. The three isolates, *Serratia marcescens*, *Streptomyces viridodiasticus* and *Micromonospora carbonacea*, significantly reduced the growth of *S. minor* in vitro, and produced high levels of chitinase and **beta-1,3-glucanase**. *Streptomyces viridodiasticus* also produced antifungal metabolite(s) that significantly reduced the growth of the pathogen in vitro. When the pathogen was presented as the sole carbon source, all three isolates caused extensive hyphal plasmolysis and cell wall lysis. *Serratia marcescens* and *St. viridodiasticus* were competent to varying degrees in colonizing the roots of lettuce seedlings after 8 days on agar plates and the rhizosphere within 14 days in pots, with their competency being superior to that of *M. carbonacea*. All three isolates, individually or in combination, were antagonistic to *S. minor* and significantly reduced incidence of disease under controlled glasshouse conditions.

L6 ANSWER 8 OF 10 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 1999:66919 BIOSIS
DOCUMENT NUMBER: PREV199900066919
TITLE: Biological control of fusarium wilt of cucumber by **chitinolytic bacteria**.
AUTHOR(S): Singh, Pushpinder Paul; Shin, Yong Chul; Park, Chang Seuk; Chung, Young Ryun [Reprint author]
CORPORATE SOURCE: Dep. Microbiol., Gyeongsang Natl. Univ., Chinju 660-701, South Korea
SOURCE: Phytopathology, (Jan., 1999) Vol. 89, No. 1, pp. 92-99. print.
CODEN: PHYTAJ. ISSN: 0031-949X.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 16 Feb 1999

Last Updated on STN: 16 Feb 1999

AB Two **chitinolytic** bacterial strains, *Paenibacillus* sp. 300 and *Streptomyces* sp. 385, suppressed *Fusarium* wilt of cucumber (*Cucumis sativus*) caused by *Fusarium oxysporum* f. sp. *cucumerinum* in nonsterile, soilless potting medium. A mixture of the two strains in a ratio of 1:1 or 4:1 gave significantly ($P < 0.05$) better control of the disease than each of the strains used individually or than mixtures in other ratios. Several formulations were tested, and a zeolite-based, chitosan-amended formulation (ZAC) provided the best protection against the disease. Dose-response studies indicated that the threshold dose of 6 g of formulation per kilogram of potting medium was required for significant ($P < 0.001$) suppression of the disease. This dose was optimum for maintaining high rhizosphere population densities of **chitinolytic bacteria** (log 8.1 to log 9.3 CFU/g dry weight of potting medium), which were required for the control of *Fusarium* wilt. The ZAC formulation was suppressive when added to pathogen-infested medium 15 days before planting cucumber seeds. The formulation also provided good control when stored for 6 months at room temperature or at 4degreeC. Chitinase and **beta-1,3-glucanase** enzymes were produced when the strains were grown in the presence of colloidal **chitin** as the sole carbon source. Partial purification of the chitinases, followed by sodium dodecyl sulfate polyacrylamide gel electrophoresis and activity staining, revealed the presence of five bands with molecular masses of 65, 62, 59, 55, and 52 kDa in the case of *Paenibacillus* sp. 300; and three bands with molecular masses of 52, 38, and 33 kDa in the case of *Streptomyces* sp. 385. Incubation of cell walls of *F. oxysporum* f. sp. *cucumerinum* with partially purified enzyme fractions led to the release of N-acetyl-D-glucosamine (NAGA). NAGA content was considerably greater when pooled enzyme fractions (64 to 67) from *Paenibacillus* sp. were used, because they contained high **beta-1,3-glucanase** activity in addition to chitinase activity. Suppression of *Fusarium* wilt of cucumber by a combination of these two **bacteria** may involve the action of these hydrolytic enzymes.

L6 ANSWER 9 OF 10 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 1992:123532 BIOSIS
DOCUMENT NUMBER: PREV199293069332; BA93:69332
TITLE: EVIDENCE THAT CHITINASE PRODUCED BY AEROMONAS-CAVIAE IS INVOLVED IN THE BIOLOGICAL CONTROL OF SOIL-BORNE PLANT PATHOGENS BY THIS BACTERIUM.
AUTHOR(S): INBAR J [Reprint author]; CHET I
CORPORATE SOURCE: DEP PLANT PATHOLOGY MICROBIOL, FAC AGRIC, HEBREW UNIV JERUSALEM, REHOVOT 76100, ISRAEL
SOURCE: Soil Biology and Biochemistry, (1991) Vol. 23, No. 10, pp. 973-978.
CODEN: SBIOAH. ISSN: 0038-0717.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 1 Mar 1992
Last Updated on STN: 2 Mar 1992

AB A **chitinolytic** isolate of *Aeromonas caviae* was isolated from roots of healthy bean plants grown in soil artificially infested with *Sclerotium rolfsii*. Under greenhouse conditions, the bacterium controlled *Rhizoctonia solani* and *Fusarium oxysporum* f.sp. *vasinfectum* in cotton (78 and 57% disease reduction, respectively) and *S. rolfsii* in beans (60% disease reduction). Seed coating was the most effective application method for controlling *R. solani* in cotton. There was no evidence of inhibition of the fungal pathogens by *A. caviae*. *A. caviae* partially lysed live mycelium of *R. solani*, *S. rolfsii* and *F. oxysporum* f.sp. *vasinfectum* when their mycelium served as a sole carbon source in liquid medium. A high **chitinolytic** activity was found when colloidal **chitin** was used as a sole carbon source, with an optimum pH between 6.0-7.0. No **beta-1-3-glucanase** was produced by the bacterium. After partial purification of the enzyme by affinity adsorption to colloidal **chitin**, three bands appear in sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). One strong band with a

molecular weight of ca. 80 kDa, and two weak bands with molecular weights of 48 and 59 kDa. Using the chromogenic substrate pNp-chitobiose, the partially purified chitinase from *A. caviae* was shown to act in an exo-splitting manner.

L6 ANSWER 10 OF 10 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1983:294828 BIOSIS
DOCUMENT NUMBER: PREV198376052320; BA76:52320
TITLE: CHITINASE IN BEAN PHASEOLUS-VULGARIS LEAVES INDUCTION BY ETHYLENE PURIFICATION PROPERTIES AND POSSIBLE FUNCTION.
AUTHOR(S): BOLLER T [Reprint author]; GEHRI A; MAUCH F; VOGELI U
CORPORATE SOURCE: BOTANISCHES INST UNIV, SCHOENBEINSTR 6, CH-4056 BASEL, SWITZERLAND
SOURCE: Planta (Heidelberg), (1983) Vol. 157, No. 1, pp. 22-31.
CODEN: PLANAB. ISSN: 0032-0935.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH

AB Ethylene induced an endochitinase in primary leaves of *P. vulgaris* L. The enzyme formed chitobiose and higher **chitin** oligosaccharides from insoluble, colloidal or regenerated **chitin**. Less than 5% of the total **chitinolytic** activity was detected in an exochitinase assay proposed by Abeles et al. (1970) for ethylene-induced chitinase. In ethylene-treated plants, chitinase activity started to increase after a lag of 6 h and was induced 30-fold within 24 h. Exogenously supplied ethylene at 1 nl ml⁻¹ was sufficient for half-maximal induction, and enhancement of the endogenous ethylene formation also enhanced chitinase activity. Cycloheximide prevented the induction. Among various hydrolases tested, only chitinase and, to a lesser extent, **beta** **-1,3-glucanase** were induced by ethylene. Induction of chitinase by ethylene occurred in many different plant species. Ethylene-induced chitinase was purified by affinity chromatography on a column of regenerated **chitin**. Its apparent MW obtained by sodium dodecyl sulfate-gel electrophoresis was 30,000; the MW determined from filtration through Sephadex G-75 was 22,000. The purified enzyme attacked **chitin** in isolated cell walls of *Fusarium solani*. It also acted as a lysozyme when incubated with *Micrococcus lysodeikticus*. Apparently, ethylene-induced chitinase functions as a defense enzyme against fungal and bacterial invaders.

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(FILE 'HOME' ENTERED AT 11:23:12 ON 10 MAR 2005)

FILE 'STNGUIDE' ENTERED AT 11:23:19 ON 10 MAR 2005

FILE 'HOME' ENTERED AT 11:23:26 ON 10 MAR 2005

FILE 'MEDLINE, HCAPLUS, BIOSIS, BIOTECHDS, EMBASE, SCISEARCH' ENTERED AT 11:24:04 ON 10 MAR 2005

L1 1 S N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGUS
L2 1 S N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGI
L3 708 S CHITIN AND BACTERIA AND FUNGI
L4 531 DUP REM L3 (177 DUPLICATES REMOVED)
L5 69 S L4 AND CHITINOLYTIC
L6 10 S L5 AND (ACETYLHEXOSAMINIDASE OR BETA-1 3 GLUCANASE)

=> s 15 and acetylglucosamine

L7 8 L5 AND ACETYLGLUCOSAMINE

=> d 17 1-8 ibib ab

L7 ANSWER 1 OF 8 MEDLINE on STN
ACCESSION NUMBER: 2001424893 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11472904
TITLE: Growth of **chitinolytic** dune soil beta-subclass Proteobacteria in response to invading fungal hyphae.

AUTHOR: De Boer W; Klein Gunnewiek P J; Kowalchuk G A; Van Veen J A
 CORPORATE SOURCE: Netherlands Institute of Ecology, Centre for Terrestrial Ecology, Department of Plant-Microorganism Interactions, 6666 ZG Heteren, The Netherlands.. wdeboer@cto.nioo.knaw.nl
 SOURCE: Applied and environmental microbiology, (2001 Aug) 67 (8) 3358-62.
 Journal code: 7605801. ISSN: 0099-2240.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 OTHER SOURCE: GENBANK-AJ310394; GENBANK-AJ310395
 ENTRY MONTH: 200110
 ENTRY DATE: Entered STN: 20011029
 Last Updated on STN: 20011029
 Entered Medline: 20011025

AB It has frequently been reported that **chitinolytic** soil **bacteria**, in particular biocontrol strains, can lyse living fungal hyphae, thereby releasing potential growth substrate. However, the conditions used in such assays (high bacterial density, rich media, fragmented hyphae) make it difficult to determine whether mycolytic activity is actually of importance for the growth and survival of **chitinolytic bacteria** in soils. An unidentified group of beta-subclass Proteobacteria (CbetaPs) was most dominant among the culturable nonfilamentous **chitinolytic bacteria** isolated from Dutch sand dune soils. Here we demonstrate that the CbetaPs grew at the expense of extending fungal mycelium of three dune soil **fungi** (*Chaetomium globosum*, *Fusarium culmorum*, and *Mucor hiemalis*) under nutrient-limiting, soil-like conditions. Aggregates of CbetaPs were also often found attached to fungal hyphae. The growth of a control group of dominant nonchitinolytic dune soil **bacteria** (beta- and gamma-subclass Proteobacteria) was not stimulated in the mycelial zone, indicating that growth-supporting materials were not independently released in appreciable amounts by the extending hyphae. Therefore, mycolytic activities of CbetaPs have apparently been involved in allowing them to grow after exposure to living hyphae. The chitinase inhibitor allosamidin did not, in the case of *Mucor*, or only partially, in the cases of *Chaetomium* and *Fusarium*, repress mycolytic growth of the CbetaPs, indicating that chitinase activity alone could not explain the extent of bacterial proliferation. **Chitinolytic** *Stenotrophomonas*-like and *Cytophaga*-like **bacteria**, isolated from the same dune soils, were only slightly stimulated by exposure to fungal hyphae.

L7 ANSWER 2 OF 8 MEDLINE on STN
 ACCESSION NUMBER: 93307280 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 8319677
 TITLE: Characteristics of an exochitinase from *Streptomyces olivaceoviridis*, its corresponding gene, putative protein domains and relationship to other chitinases.
 AUTHOR: Blaak H; Schnellmann J; Walter S; Henrissat B; Schrempf H
 CORPORATE SOURCE: FB Biologie/Chemie, University of Osnabruck, Germany.
 SOURCE: European journal of biochemistry / FEBS, (1993 Jun 15) 214 (3) 659-69.
 Journal code: 0107600. ISSN: 0014-2956.
 PUB. COUNTRY: GERMANY: Germany, Federal Republic of
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 OTHER SOURCE: GENBANK-L09682; GENBANK-L09683; GENBANK-X71080; GENBANK-X71335; GENBANK-X71336; GENBANK-X71337; GENBANK-X71338; GENBANK-X71339; GENBANK-X71340; GENBANK-X72968
 ENTRY MONTH: 199308
 ENTRY DATE: Entered STN: 19930813
 Last Updated on STN: 19950206
 Entered Medline: 19930803

AB *Streptomyces olivaceoviridis* efficiently degrades **chitin**. Shotgun cloning of partially Sau3A-cleaved DNA using the multicopy vector pIJ702 and *Streptomyces lividans* 66 as host resulted in the identification

of the plasmid pCHI 01 which harbours an insert of 4.6 kb. In the presence of **chitin** as sole carbon source, transformants of *S. lividans* 66 carrying pCHI 01 or its derivatives with smaller inserts overproduced an exochitinase which was purified to homogeneity. The **chitin**-inducible enzyme with an isoelectric point of 4.0 shows optimal activity at pH 7.3 and 55 degrees C, has an apparent molecular mass of 47 kDa and is competitively inhibited by the pseudosugar allosamidin. The enzyme was identified as an exochitinase since it generates exclusively chitobiose from chitotetraose, chitohexaose, and colloidal high-molecular mass **chitin**. Sequence analysis of a reading frame of 1794 base pairs and comparison of the deduced amino-acid sequence allowed the identification of the putative catalytic domain, one region with significant similarity to the type-III module of fibronectin and one domain of unknown function. Multiple sequence alignment and hydrophobic-cluster analysis of 25 **chitinolytic** enzymes from **bacteria**, **fungi** and plants allowed the identification of their characteristic domains. The exochitinase from *S. olivaceoviridis* shares highest similarity with the chitinase D from *Bacillus circulans*.

L7 ANSWER 3 OF 8 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1996:360706 HCAPLUS

DOCUMENT NUMBER: 125:28825

TITLE: Chitinolysis by the marine ascomycete *Corollospora maritima* Werdermann: purification and properties of a chitobiosidase

AUTHOR(S): Grant, W. D.; Atkinson, M.; Burke, B.; Molloy, C.

CORPORATE SOURCE: Cawthron Institute, Nelson, N. Z.

SOURCE: Botanica Marina (1996), 39(2), 177-186

CODEN: BOTNA7; ISSN: 0006-8055

PUBLISHER: de Gruyter

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Out of six marine **fungi** examd., two, *Corollospora maritima* Werdermann, strain 198 and *Lindra obtusa* Nakagiri et Tubaki, strain 4937, grew on **chitin** and produced **chitinolytic** enzymes. *Corollospora maritima* showed higher growth rates and enzyme activity and was studied further. Crude, purified and colloidal **chitin**, as well as N-acetylglucosamine (GlcNAc) and the **chitin** di- and trisaccharides (GlcNAc)₂ and (GlcNAc)₃, produced fungal growth comparable to that from carbon sources unrelated to **chitin**. Only **chitin**-related substrates induced **chitinolytic** activity. Growth in 0.2% (w/v) purified **chitin** and 0.2% (w/v) GlcNAc was maximal after 12-14 days. **Chitin**-induced mycelial and extracellular (culture fluid) **chitinolytic** activities peaked at 10 and 14 days resp., whereas the activities in both fractions increased in parallel in GlcNAc-induced cultures with twice as much activity in the culture fluid as in the mycelium throughout. **Chitin** and GlcNAc both induced **chitinolytic** activity in washed, glucose-grown mycelia but no activity appeared if glucose was present during the induction period. The culture fluid of GlcNAc-induced mycelia contained N-acetyl-.beta.-glucosaminidase activity (EC 3.2.1.30) and a **chitin** 1,4-chitobiosidase as defined previously. The latter enzyme was purified by adsorption to colloidal **chitin** and desorption by enzymic digestion of the substrate, followed by chromatofocusing. One peak of **chitinolytic** activity eluted from the chromatofocusing column at pH 7.55 and corresponded to a single polypeptide of relative mol. mass (Mr) 40 000 identified by SDS-PAGE. High performance liq. chromatog. anal. showed that the major hydrolysis product from colloidal **chitin**, purified **chitin** and the **chitin** tetrasaccharide (GlcNAc)₄ was (GlcNAc)₂ (> 90% of the reducing sugars), the remainder being GlcNAc and (GlcNAc)₃. The substrate (GlcNAc)₃ also yielded (GlcNAc)₂, indicating that the chitobiosidase catalyzes a transglycosylation reaction. This 'exochitinase' depolymd. crude **chitin** demonstrating that *Corollospora maritima* utilizes a **chitinolytic** enzyme system more akin to that of **chitinolytic bacteria** than to terrestrial **chitinolytic fungi**.

L7 ANSWER 4 OF 8 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1996:134684 HCAPLUS
 DOCUMENT NUMBER: 124:174586
 TITLE: Variability of N mineralization and nitrification in a simple, simulated microbial forest soil community
 AUTHOR(S): De Boer, W.; Gunnewiek, P. J. A. Klein; Parkinson, D.
 CORPORATE SOURCE: Netherlands Inst. Ecology, Cent. Terrestrial Ecology, Heteren, 6666 ZG, Neth.
 SOURCE: Soil Biology & Biochemistry (1996), 28(2), 203-11
 CODEN: SBIOAH; ISSN: 0038-0717
 PUBLISHER: Elsevier
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB It is generally accepted that N transformations in natural terrestrial ecosystems are regulated by (1) climatol. and geomorphol. conditions, (2) soil physics and chem., and (3) quality and quantity of soil org. matter. Consequently, spatial variability of N transformations within exptl. plots have been related to fine-scale heterogeneity of these regulating factors, e.g. spatial differences in micro-climate. However, it has also been argued that spatial differences in the compn. of the microbial community, which can be the result of stochastic events, may be an important source of spatial heterogeneity of decompn. processes. The aim of the study was to detect to what extent net N mineralization and nitrification can vary in soil samples as a result of differences in the compn. of a simple microbial community of acid forest soils. This community consisted of two species of **chitin** decomposers, a mycelium-forming fungus and a rod-like bacterium, and two species of nitrifying **bacteria**. Characterization of the **chitin** decomposers in liq. cultures demonstrated two important differences between the fungus and the bacterium: (1) **chitin**-N mineralization by the bacterium was much slower than that by the fungus, and (2) the bacterium showed an antibiotic-type of inhibition against the nitrifying **bacteria** whereas the fungus did not. The effect of differences in the compn. of the **chitinolytic** community on N mineralization and nitrification was studied using environmentally controlled incubations of the microorganisms in Petri-dishes contg. purified sand with solid **chitin** or its sol. monomer, **N-acetylglucosamine**, as substrate. As seen in liq. cultures, the differences in **chitin**-N mineralization between series of sand incubations of either the fungus or the bacterium were considerable. Surprisingly, **chitin**-N mineralization in sand that had been inoculated with both **chitinolytic** decomposers, was relatively slow. This was most likely due to mycolytic activity of the bacterium against the fungus. Variation of N mineralization within series of identical inoculation, consisting of 10 replicates, was generally low (CV <15%). In contrast, nitrification was extremely variable within many sand series (CV <200%), esp. those in which the **chitinolytic** bacterium was present. This high variability of nitrification was most likely due to instability of the antibiotic compd. in sand. In conclusion, the results show that spatial variability in the compn. of a simple microbial community and interactions therein can be an important source of small-scale heterogeneity of N transformations. The relevance of these results for the field situation is discussed.

L7 ANSWER 5 OF 8 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1995:558736 HCAPLUS
 DOCUMENT NUMBER: 122:310385
 TITLE: **Chitinolytic** Enterobacter agglomerans antagonistic to fungal plant pathogens
 AUTHOR(S): Chernin, Leonid; Ismailov, Zafar; Haran, Shoshan; Chet, Ilan
 CORPORATE SOURCE: Otto Warburg Center Biotechnology Agriculture, Fac. Agriculture, Hebrew Univ. Jerusalem, Rehovot, 76100, Israel
 SOURCE: Applied and Environmental Microbiology (1995), 61(5), 1720-6
 CODEN: AEMIDF; ISSN: 0099-2240
 PUBLISHER: American Society for Microbiology
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Three *Enterobacter* agglomerans strains which produce and excrete proteins with **chitinolytic** activity were found while screening soil-borne **bacteria** antagonistic to fungal plant pathogens. The **chitinolytic** activity was induced when the strains were grown in the presence of colloidal **chitin** as the sole carbon source. It was quantitated by using assays with chromogenic p-nitrophenyl analogs of disaccharide, trisaccharide, and tetrasaccharide derivs. of N-**acetylglucosamine**. A set of three fluorescent substrates with a 4-methylumbelliferyl group linked by .beta.-1,4 linkage to N-**acetylglucosamine** mono- or oligosaccharides were used to identify the **chitinolytic** activities of proteins which had been renatured following their sepn. by electrophoresis. This study provides the most complete evidence for the presence of a complex of **chitinolytic** enzymes in *Enterobacter* strains. Four enzymes were detected: two N-acetyl-.beta.-D-glucosaminidases of 89 and 67 kDa, an endochitinase with an apparent mol. mass of 59 kDa, and a chitobiosidase of 50 kDa. The biocontrol ability of the **chitinolytic** strains was demonstrated under greenhouse conditions. The **bacteria** decreased the incidence of disease caused by *Rhizoctonia solani* in cotton by 64 to 86%. Two Tn5 mutants of one of the isolates, which were deficient in **chitinolytic** activity, were unable to protect plants against the disease.

L7 ANSWER 6 OF 8 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 2004:452162 BIOSIS
DOCUMENT NUMBER: PREV200400456577
TITLE: Production dynamics and characterization of
chitinolytic system of *Streptomyces* sp. NK1057, a
well equipped **chitin** degrader.
AUTHOR(S): Nawani, N. N.; Kapadnis, B. P. [Reprint Author]
CORPORATE SOURCE: Dept Microbiol, Univ Poona, Poona, Maharashtra, 411007,
India
bpkapadnis@yahoo.com
SOURCE: World Journal of Microbiology & Biotechnology, (July 2004)
Vol. 20, No. 5, pp. 487-494. print.
ISSN: 0959-3993 (ISSN print).
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 24 Nov 2004
Last Updated on STN: 24 Nov 2004

AB *Streptomyces* sp. NK1057 produced four extracellular chitinases. Two (62 and 48 kDa) were endochitinases while the other two (35 and 28 kDa) had chitobiosidase activity. Chi62 was produced in early growth phase whereas the other three were produced later. Chi48 was optimally active at pH 4.0 and 60degreeC whereas Chi35 was optimally active at pH 6.0 and 40degreeC. Both the enzymes had fairly good pH and temperature stability up to 50degreeC, with Chi48 being more thermostable than Chi35. Chi48 was significantly inhibited by N-**acetylglucosamine** but not by Hg2+ and the reverse was true for Chi35. Chi48 and Chi35 had isoelectric points of 5.1 and 6.0 and the N-terminal amino acid sequence of Chi35 determined up to four amino acids, was Gln-Ser-Pro-Gly. Chi62 and Chi48 were able to inhibit the germination of *Fusarium oxysporum* spores by 57.4 and 61.2% respectively whereas; Chi35 and Chi28 did so by only 14.1 and 3.8%, suggesting endochitinase to possess antifungal activity. Only Chi28 exhibited a lytic activity, 0.022 U ml-1, towards *Micrococcus lysodeikticus* cells.

L7 ANSWER 7 OF 8 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 1996:67058 BIOSIS
DOCUMENT NUMBER: PREV199698639193
TITLE: Enzyme formation by a yeast cell wall lytic *Arthrobacter*
species: **Chitinolytic** activity.
AUTHOR(S): Latzko, F.; Hampel, W. [Reprint author]
CORPORATE SOURCE: Inst. Biochem. Technol. Microbiol., Vienna Univ. Technol.,
Getreidemarkt 9/172, A-1060 Vienna, Austria
SOURCE: Applied Microbiology and Biotechnology, (1995) Vol. 44, No.
1-2, pp. 185-189.
CODEN: AMBIDG. ISSN: 0175-7598.
DOCUMENT TYPE: Article

LANGUAGE: English
ENTRY DATE: Entered STN: 9 Feb 1996
Last Updated on STN: 10 Feb 1996

AB The kinetics of the release of **chitinolytic** activity (endochitinase EC 3.2.1.14, beta-N-acetylglucosaminidase EC 3.2.1.30) by a yeast cell wall lytic *Arthrobacter* species was studied. The organism was cultivated on yeast cell wall, mycelium of *Trichoderma reesei*, colloidal **chitin**, **N-acetylglucosamine**, glucosamine and mixtures with acetate. With the exception of yeast cell wall, these substrates were used as the sole source of carbon and nitrogen. The growth on colloidal **chitin** (0.5%) proceeded at a maximum specific growth rate (μ -max) of 0.23 h⁻¹ and yielded 2700 mU l⁻¹ chitinase. Yeast cell wall and mycelium of *T. reesei* supported more rapid growth (μ -max = 0.30 h⁻¹ and 0.25 h⁻¹ respectively) but yielded reduced chitinase activity (565 mU l⁻¹ and 760 mU l⁻¹). The growth rate on glucosamine (μ -max = 0.24 h⁻¹) was reduced when this was mixed with acetate (μ -max = 0.12 h⁻¹), whereas the enzyme yield was increased from 720 mU l⁻¹ to 960 mU l⁻¹. The same effect on growth rate was observed with glucose and equimolar mixtures of glucose and acetate, indicating a strong impact of the organic acid on carbohydrate transport or metabolism. The growth of adapted cells on **N-acetylglucosamine** was comparable to that observed on an equimolar mixture of glucosamine and acetate, indicating that **N-acetylglucosamine** is rapidly hydrolysed by adapted cells.

L7 ANSWER 8 OF 8 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 1987-13897 BIOTECHDS

TITLE: Distribution of **chitinolytic** activity in *Bacillus* species;
Bacillus thuringiensis, *Bacillus alvei*, *Bacillus laterosporus*, *Bacillus coagulans* (conference abstract)

AUTHOR: Cody R M

LOCATION: Botany and Microbiology, Auburn University, Auburn, Alabama, USA.

SOURCE: Abstr. Annu. Meet. Am. Soc. Microbiol.; (1987) 87 Meet., 294

DOCUMENT TYPE: Journal

LANGUAGE: English

AB **Chitin**, a polymer of beta-1,4-N-acetylglucosamine is degraded primarily by **bacteria** and **fungi**. Actinomycetes represent 90-99% of the chitinoclastic organisms from arable soils. Other **chitinolytic bacteria** are represented by *Bacillus*, *Myxobacter*, *Serratia* spp., etc. An examination of 20 laboratory reference cultures and over 200 field isolates of *Bacillus* by the Berger clear zone-colony ratio method and measurement of reducing sugar with 3,5-dinitrosalicylic acid reagent showed that **chitinolytic** activity is limited to a few spp. The most prominent **chitinolytic** spp. are *Bacillus alvei*, *Bacillus laterosporus*, *Bacillus thuringiensis* and field isolates tentatively identified as *Bacillus coagulans*. **Chitinolytic** activity of all spp. and isolates was increased by incorporating yeast extract in Bushnell-Hass agar. (0 ref)

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FILE 'STNGUIDE' ENTERED AT 11:23:19 ON 10 MAR 2005

FILE 'HOME' ENTERED AT 11:23:26 ON 10 MAR 2005

FILE 'MEDLINE, HCAPLUS, BIOSIS, BIOTECHDS, EMBASE, SCISEARCH' ENTERED AT 11:24:04 ON 10 MAR 2005

L1 1 S N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGUS
L2 1 S N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGI
L3 708 S CHITIN AND BACTERIA AND FUNGI
L4 531 DUP REM L3 (177 DUPLICATES REMOVED)
L5 69 S L4 AND CHITINOLYTIC
L6 10 S L5 AND (ACETYLHEXOSAMINIDASE OR BETA-1 3 GLUCANASE)
L7 8 S L5 AND ACETYLGLUCOSAMINE

=> s N-acetylglucosamine formation and chitinase?
L8 4 N-ACETYLGLUCOSAMINE FORMATION AND CHITINASE?

=> dup rem l8
PROCESSING COMPLETED FOR L8
L9 4 DUP REM L8 (0 DUPLICATES REMOVED)

=> d l9 1-4 ibib ab

L9 ANSWER 1 OF 4 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 2003:363761 BIOSIS
DOCUMENT NUMBER: PREV200300363761
TITLE: Saccharification of chitin using solid-state culture of
Aspergillus sp. S1-13 with shellfish waste as a substrate.
AUTHOR(S): Rattanakit, Nopakarn; Yano, Shigekazu; Wakayama, Mamoru;
Plikomol, Abhinya; Tachiki, Takashi [Reprint Author]
CORPORATE SOURCE: Department of Bioscience and Biotechnology, Faculty of
Science and Engineering, Ritsumeikan University, Kusatsu,
Shiga, 525-8577, Japan
tachiki@se.ritsumei.ac.jp
SOURCE: Journal of Bioscience and Bioengineering, (2003) Vol. 95,
No. 4, pp. 391-396. print.
ISSN: 1389-1723.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 6 Aug 2003
Last Updated on STN: 6 Aug 2003

AB Saccharification of chitin was performed in a suspension (mash) of a
solid-state culture of **chitinase**-producing *Aspergillus* sp.
S1-13 with acid-treated shellfish waste as a substrate. The conditions
for the saccharifying reaction and the solid-state cultivation were
examined from the viewpoint of saccharification in the mash. Optimum
cultivation conditions were defined: a solid-state medium consisting of 5
g of 10% lactic acid-treated crab shells (0.50-2.36 mm in size) and 3 ml
of a basal medium (0.028% KH₂PO₄, 0.007% CaCl₂·2H₂O, and 0.025%
MgSO₄·7H₂O) supplemented with 0.3% peptone was inoculated with 4 ml
of spore suspension (1×10⁷ spores/ml), and the water content of the medium
was adjusted to 75%; static cultivation at 37°C for 7 d. When a
culture obtained under the optimum conditions was suspended in 70 ml of 50
mM sodium phosphate-citrate buffer (pH 4.0) and incubated at 45°C for
11-13 d, 55 mM N-acetylglucosamine (GlcNAc) was formed in the solid-state
culture mash, indicating that at least 33% of the initial chitin in the
solid material was hydrolyzed. Through the experiments, the amounts of
GlcNAc formed in the solid-state culture mash varied in a way similar to
that of the water-extractable p-nitrophenyl beta-D-N-acetylglucosaminide-
hydrolyzing enzyme in the culture, but not to that of the colloidal
chitin-hydrolyzing enzyme. GlcNAc-assimilating lactic acid bacteria,
which were inoculated into the mash after or at the start of the
saccharification, formed lactic acid with decreasing GlcNAc.

L9 ANSWER 2 OF 4 HCAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 1992:209702 HCAPLUS
DOCUMENT NUMBER: 116:209702
TITLE: Control of *Fusarium* infections in plants by
chitinase-producing microorganisms
INVENTOR(S): Toyoda, Hideyoshi; Omoto, Masaaki
PATENT ASSIGNEE(S): Coop Chemical Co., Ltd., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 4 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 04001109	A2	19920106	JP 1990-101560	19900419
JP 2915960	B2	19990705		

AB Plant infections by *Fusarium* are controlled by chitin degradn. products of **chitinase**-forming microorganisms, such as *Serratia* and *Streptomyces*, that live on the surface of roots in the presence of chitin and chitin-contg. substances in soils. The enzyme degraded chitin to chitotriose and chitopentose, producing N-acetylglucosamine as the final product. *Fusarium* infections in tomato, cucumber, strawberry, cabbage, etc. were inhibited by chitin degradn. products by *Serratia*.

L9 ANSWER 3 OF 4 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1992:422226 HCAPLUS

DOCUMENT NUMBER: 117:22226

TITLE: Purification and characterization of a novel type of **chitinase** from *Vibrio alginolyticus* TK-22

AUTHOR(S): Murao, Sawao; Kawada, Teruhiko; Itoh, Homare; Oyama, Hiroshi; Shin, Takashi

CORPORATE SOURCE: Dep. Appl. Microb. Technol., Kumamoto Inst. Technol., Kumamoto, 860, Japan

SOURCE: Bioscience, Biotechnology, and Biochemistry (1992), 56(2), 368-9

CODEN: BBBIEJ; ISSN: 0916-8451

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A novel **chitinase** was found in the culture filtrate of *V. alginolyticus* TK-22. Partial purifn. yielded 4 **chitinase** activities. **Chitinase** A, which showed a unique substrate specificity, was further purified. The mol. wt. of the enzyme was estd. to be 66,000 by SDS-PAGE, and 57,000 by gel filtration. The pI was 4.3 and the E1%1 cm, 280nm value was 11.5. The optimum pH for the enzyme activity was unique, showing 2 optima at pH 4 and 9. The temp. optimum was 45.degree.. The reaction products from colloidal chitin were N-acetylglucosamine, chitobiose, chitotriose, chitotetraose, and chitopentaose.

L9 ANSWER 4 OF 4 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1993:513086 HCAPLUS

DOCUMENT NUMBER: 119:113086

TITLE: Degradation pathway of chitin by a thermophilic bacterium *Bacillus licheniformis* X-7u

AUTHOR(S): Shimahara, Kenzo; Takiguchi, Yasuyuki; Nagano, Yoko

CORPORATE SOURCE: Fac. Eng., Seikei Univ., Musashino, 180, Japan

SOURCE: Adv. Chitin Chitosan, [Proc. Int. Conf.], 5th (1992), Meeting Date 1991, 364-71. Editor(s): Brine, Charles J.; Sanford, Paul A.; Zikakis, John P. Elsevier: London, UK.

CODEN: 58YVAW

DOCUMENT TYPE: Conference

LANGUAGE: English

AB **Chitinase** and N-acetyl-.beta.-D-glucosaminidase (GlcNAcase) were both induced in a culture fluid of X-7u grown in a medium contg. colloidal chitin. Both enzymes were also produced by Di-N-acetylchitobiose (GlcNAc(2)). During the cultivation, N-acetylglucosamine (GlcNAc) was produced by 2 pathways: by direct hydrolysis of chitin catalyzed by **chitinase** to produce (GlcNAc)2 and GlcNAc; and by the (GlcNAc)2 catalyzed by both **chitinase** and GlcNAcase. The GlcNAc produced was immediately incorporated into the X-7u cells and thus did not accumulate in the culture fluid. On the basis of these and other results, a scheme of the degradn. pathway of chitin was prepd.

=> s N-acetylglucosamine and (trichoderma or aphanocladium or coccidioides or aspergillus or hyp
L10 69 N-ACETYLGLUCOSAMINE AND (TRICHODERMA OR APHANOCLADIUM OR COCCIDI
OIDES OR ASPERGILLUS OR HYPOCREA OR CANDIDA) AND (AEROMONAS OR
STREPTOMYCES OR SERRATIA OR BACILLUS OR CHROMOBACTER OR VIBRIO
OR PSEUDOMONAS OR PYROCOCCUS)

=> dup rem 110

PROCESSING COMPLETED FOR L10

L11 37 DUP REM L10 (32 DUPLICATES REMOVED)

=> focus l11
PROCESSING COMPLETED FOR L11
L12. 37 FOCUS L11 1-

=> d 112 1-37 ibib ab

L12 ANSWER 1 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2004:20837 HCAPLUS
DOCUMENT NUMBER: 140:92684
TITLE: Genetically engineered Escherichia coli strains and
fermentation process for production of glucosamine and
N-acetylglucosamine
INVENTOR(S): Deng, Ming-de; Angerer, J. David; Cyron, Don; Grund,
Alan D.; Jerrell, Thomas A., Jr.; Leanna, Candice;
Mathre, Owen.; Rosson, Reinhardt; Running, Jeff;
Severson, Dave; Song, Linsheng; Wassink, Sarah
PATENT ASSIGNEE(S): Arkion Life Sciences Llc, USA
SOURCE: PCT Int. Appl., 327 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004003175	A2	20040108	WO 2003-US20925	20030701
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW			
RW:	GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG			
US 2004091976	A1	20040513	US 2003-612779	20030701
PRIORITY APPLN. INFO.:			US 2002-393348P	P 20020701

OTHER SOURCE(S): CASREACT 140:92684

AB A biosynthetic method for producing glucosamine and **N-acetylglucosamine** is disclosed. Such a method includes the fermn. of a genetically modified microorganism to produce glucosamine and/or **N-acetylglucosamine**. Also disclosed are genetically modified microorganisms that are useful for producing glucosamine and **N-acetylglucosamine**. In addn., methods of recovering **N-acetylglucosamine** that has been produced by a fermn. process, including methods that result in **N-acetylglucosamine** of high purity, are described. Also disclosed is a method to produce glucosamine from **N-acetylglucosamine**.

L12 ANSWER 2 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2002:142910 HCAPLUS
DOCUMENT NUMBER: 136:199030
TITLE: Production and use of inducible enzymes from
Trichoderma and bacteria for control of plant
pests and for industrial processes
INVENTOR(S): Harman, Gary E.; Donzelli, Bruno; Deng, Shiping
PATENT ASSIGNEE(S): Cornell Research Foundation, Inc., USA
SOURCE: PCT Int. Appl., 53 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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 WO 2002014540 A1 20020221 WO 2001-US25058 20010810
 W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
 CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR,
 HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT,
 LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU,
 SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU,
 ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
 RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,
 DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF,
 BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG
 AU 2001084793 A5 20020225 AU 2001-84793 20010810
 US 2004265953 A1 20041230 US 2001-927984 20010810
 PRIORITY APPLN. INFO.: US 2000-225700P P 20000811
 WO 2001-US25058 W 20010810

AB The present invention relates to a method of increasing the expression of native extracellular inducible microbial enzymes from fungal sources using particular culture media and a method of repressing native proteins in fungi and enhancing the prodn. of proteins encoded by transgenes in fungi under the control of an inducible promoter. The present invention also relates to mixts. of extracellular inducible enzymes from a fungal and a bacterial source as well as their use in a method for the degrdn. of chitinous substrates. Methods of releasing **N-acetylglucosamine** from chitinous sources and enhancing purity of a heterologous recombinant protein expressed in a culture of fungi are also disclosed.

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 3 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1958:84422 HCAPLUS
 DOCUMENT NUMBER: 52:84422
 ORIGINAL REFERENCE NO.: 52:14923g-i,14924a-d
 TITLE: Aerobic decomposition of chitin by microorganisms
 AUTHOR(S): Veldkamp, H.
 SOURCE: Mededel. Landouwhogeschool Wageningen (1955), 55, 127-74
 From: Biol. Abstr. 31, Abstr. No. 22036(1957)
 DOCUMENT TYPE: Journal
 LANGUAGE: Unavailable

AB Strains (50) of chitin-decomp. bacteria belonging to *Achromobacter*, *Flavobacterium*, *Chromobacterium*, ***Bacillus***, *Cytophaga*, and ***Pseudomonas***, 23 chitinovorous actinomycetes (***Streptomyces***, *Micromonospora*, and *Nocardia*), and several chitin-decomp. fungi (one could be identified as ***Aspergillus*** *fumigatus* and the others as members of *Mortierella*) were isolated from soil. In all soils, actinomycetes formed the major part of the population of chitin-decomp. microorganisms, whereas the per cent of actinomycetes in "total" microorganisms was relatively small; bacteria formed 1-3% and fungi less than 1%. In soils satd. with water, only chitinovorous bacteria achieve a rapid multiplication, whereas actinomycetes do not develop in such soils. In all soils to which chitin was added, the changes in the microbial population resulted in a predominance of actinomycetes over bacteria. Observations of nitrification as a result of chitin-decompn. in different soils are described. A relatively rapid accumulation of nitrate could generally be seen up to about 3 weeks after chitin addn. to water-satd. or relatively dry soils. Up to 60% of the N originally present in chitin was recovered as nitrate. Investigations on hydrolysis of chitin by bacterial enzymes were carried out with ***Pseudomonas*** chitinovorans, strain 8500, and *Cytophaga johnsonae*. In cultures of both strains, contg. chitin as the sole course of N and C, **N-acetylglucosamine** as well as glucosamine could be traced. AcOH was found during chitin decompn. by both strains, indicating that **N-acetylglucosamine** is deacetylated to yield glucosamine. The aerobic breakdown of chitin and glucose by *P. chitonovorans* strain 8676 is described. This strain forms more acid from chitin than does *P. chitonovorans*, strain 8500. In a medium contg. 0.35% (NH₄)₂SO₄, the max. no. of viable cells was seen at the moment of complete disappearance of glucose. During the period of logarithmic growth, lactic acid and pyruvic

acid were found in the medium; these acids were eventually consumed. In a medium contg. 0.1% (NH₄)₂SO₄, the N supply becomes the limiting factor for growth 12 hrs. after inoculation. The cessation of bacterial multiplication was coincident with a continued production of pyruvic acid and a sudden production of .alpha.-ketoglutaric acid. As contrasted with pyruvic acid, .alpha.-ketoglutaric acid was not used further by the bacterial cells. C balances of both expts. with glucose as the substrate showed that the major part of the C recovered at the end of the expts. could be found in cell material and CO₂. A small amt. of acetoin was present in the culture liquids at the end of the expts. It is proposed that the formation of pyruvic acid from chitin depends on the concn. of the products of hydrolysis in the medium. C balances made in expts. with chitin as the substrate showed that the major part of the C initially present in chitin could be recovered as cell material and CO₂ at the end of the expts. When **N-acetylglucosamine** was used as the substrate, production of pyruvic acid was always seen. Although it is still uncertain whether glucose is an intermediate in the breakdown of **N-acetylglucosamine**, the formation of pyruvic acid from both substances by *P. chitinovorans* strain 8676 indicates that the terminal respiration pattern is the same for both substrates.

L12 ANSWER 4 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1986:125548 HCAPLUS
DOCUMENT NUMBER: 104:125548
TITLE: The chitinase system of **Streptomyces** sp.
ATCC 11238 and its significance for fungal cell wall degradation
AUTHOR(S): Beyer, M.; Diekmann, H.
CORPORATE SOURCE: Inst. Mikrobiol., Univ. Hannover, Hannover, D-3000/1, Fed. Rep. Ger.
SOURCE: Applied Microbiology and Biotechnology (1985), 23(2), 140-6
CODEN: AMBIDG; ISSN: 0175-7598
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A crude prepn. of extracellular proteins from **Streptomyces** species ATCC 11238, contg. chitin and laminarin-degrading enzymes, was active in lysing the cell walls of most of 50 viable filamentous ascomycetes tested, but was almost inactive with endomycetidae, zygomycetes, and oomycetes. This mycolase prepn. was fractionated by gel filtration and DEAE-ion exchange chromatog. with special interest in chitin-degrading enzymes. **N-Acetylglucosamine** is liberated from crab shell chitin by the combined action of an exochitinase and .beta.-N-acetylglucosaminidase. Both purified enzymes lysed cell wall preps. singly or together only when supplemented by protein contg. endochitinase activity recovered from the gel after gel electrophoresis. Furthermore, enzymes degrading chitosan and azocoll were detected and sepd.

L12 ANSWER 5 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1990:6094 HCAPLUS
DOCUMENT NUMBER: 112:6094
TITLE: Manufacture of Humicola lanuginosa lipase by recombinant **Aspergillus**
INVENTOR(S): Boel, Esper; Høge-Jensen, Ida Birgitte
PATENT ASSIGNEE(S): Novo Industri A/S, Den.
SOURCE: Eur. Pat. Appl., 28 pp.
CODEN: EPXXDW
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 3
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
EP 305216	A1	19890301	EP 1988-307980	19880826
EP 305216	B1	19950802		
R: AT, BE, CH, DE, ES, FR, GB, GR, IT, LI, LU, NL, SE				
DK 8804760	A	19890419	DK 1988-4760	19880826

DK 165640	B	19921228		
DK 165640	C	19930601		
ES 2076939	T3	19951116	ES 1988-307980	19880826
JP 01157383	A2	19890620	JP 1988-212641	19880829
JP 04038394	B4	19920624		
DK 9101775	A	19911025	DK 1991-1775	19911025
US 5766912	A	19980616	US 1994-230170	19940420
US 5965384	A	19991012	US 1995-463172	19950605
US 5874558	A	19990223	US 1996-650086	19960517

PRIORITY APPLN. INFO.:

DK 1987-4500	A	19870828
DK 1987-6560	A	19871215
DK 1988-2054	A	19880415
DK 1986-1226	A	19860317
US 1987-24342	B2	19870310
US 1988-236605	B1	19880825
US 1992-954371	B3	19920930
US 1994-230170	A3	19940420
US 1995-435557	B3	19950505

AB Humicola lanuginosa lipase (I) is manufd. by culturing a transformed **Aspergillus** host, e.g. *A. niger*, *A. oryzae*, and recovering I from the culture medium. Recombinant I has better thermostability than comparable native lipase (II), is more resistant to proteolytic degrading than II, and has a different pattern of glycosylation from II. Plasmid p960 contg. the TAKA-amylase promoter from *A. oryzae*, I gene from *Humicola lanuginosa*, and AMG terminator from *A. niger* was constructed and transformed into *A. oryzae* IFO 4177 by cotransformation with P3SR2 contg. the *amdS* gene from *A. nidulans*. The *A. oryzae* transformants were cultured in 40% soybean meal plus glucose. I was recovered from the culture medium by ultrafiltration and freeze drying. I contained **N-acetylglucosamine** 1.2, mannose 8.6, and galactose 3.3 mol/mol I compared to 1.2, 5.7, and 0 mol/mol II, resp. I had better thermostability than II at pH 5-10 at 55.degree. and 60.degree., resp. and I was less susceptible to **Bacillus** protease than II.

L12 ANSWER 6 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2000:242239 HCAPLUS

DOCUMENT NUMBER: 133:160437

TITLE: Cloning, nucleotide sequence and expression of the .beta.-N-acetyl-glucosaminidase gene from **Aeromonas** sp. no. 10S-24

AUTHOR(S): Ueda, Mitsuhiro; Fujita, Yuko; Kawaguchi, Takashi; Arai, Motoo

CORPORATE SOURCE: Department of Applied Biological Chemistry, College of Agriculture, Osaka Prefecture University, Osaka, 599-8531, Japan

SOURCE: Journal of Bioscience and Bioengineering (2000), 89(2), 164-169

CODEN: JBBIF6; ISSN: 1389-1723

PUBLISHER: Society for Bioscience and Bioengineering, Japan

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The .beta.-GlcNAcase gene of **Aeromonas** sp. no. 10S-24 was cloned and sequenced. The gene consisted of 2505 bp encoding 835 amino acid residues. The amino acid sequence of the gene shares sequence homol. with .beta.-GlcNAcases of **Vibrio vulnificus** (37.4%), **Serratia marcescens** (36.4%), *V. harveyi* (36.4%), *V. parahaemolyticus* (33.5%), *Alteromonas* sp. strain O-7 (27.0%), and **Candida albicans** (28.6%). The .beta.-GlcNAcase gene of **Aeromonas** sp. no. 10S-24 was about 630-fold overexpressed in *Escherichia coli* compared to that in **Aeromonas** sp. no. 10S-24. The cloned .beta.-GlcNAcase had almost the same enzymic properties as .beta.-GlcNAcase from no. 10S-24. The carbohydrate chain of .beta.-GlcNAcase from **Aeromonas** sp. no. 10S-24 was found to contain mannose, galactose, fucose, **N-acetylglucosamine**, N-acetylgalactosamine and several other unknown saccharides.

REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 7 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1990:476557 HCAPLUS
DOCUMENT NUMBER: 113:76557
TITLE: Screening microorganisms for chitin hydrolysis and
production of ethanol from amino sugars
AUTHOR(S): Cody, R. M.; Davis, N. D.; Lin, J.; Shaw, D.
CORPORATE SOURCE: Coll. Sci. Math., Auburn Univ., Auburn, AL,
36849-5407, USA
SOURCE: Biomass (1990), 21(4), 285-95
CODEN: BIOME9; ISSN: 0144-4565
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Seventy-two strains of bacteria representing 39 genera and 1 yeast (**Candida albicans**) were screened for ability to hydrolyze chitin. Chitin hydrolysis was detd. by a clear zone surrounding colonies growing on the surface of chitin agar. Species with the largest clear zone to colony size (CZ/CS) ratio were further compared for chitinolysis by assaying the level of reducing sugar produced in broth culture. Three yeasts and 1 bacterial strain known to produce EtOH from glucose were compared for their abilities to produce EtOH from amino sugars. Of the 72 strains screened, 23 produced CZ/CS ratios of 0.38-2.5. The highest ratios were obsd. for strains of **Bacillus** and **Serratia**, followed by **Micrococcus**, **Aeromonas**, **Vibrio**, **Clostridium**, and **Plesiomonas**. The other spp. examd. produced ratios of <1 or were unable to hydrolyze chitin. **Hansenula anomala**, **Pachysolen tannophilus**, **Saccharomyces cerevisiae**, and **Zymomonas mobilis** were compared for their abilities to grow on and produce EtOH from glucose, glucosamine, and **N-acetylglucosamine** (NAG). **Saccharomyces cerevisiae** and **H. anomala** produced EtOH only from glucose. **Pachysolen tannophilus** and **Z. mobilis** produced EtOH from glucose, glucosamine, and NAG. The highest concn. of EtOH produced from amino sugar was 598 .mu.g/mL from 10 mg glucosamine/mL by **Z. mobilis**. This level was achieved only when yeast ext. was included in the medium. **Saccharomyces cerevisiae** did not grow on glucosamine and **A. mobilis** did not grow well on NAG.

L12 ANSWER 8 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2004:123941 HCAPLUS
DOCUMENT NUMBER: 140:390346
TITLE: Chitinase production by endophytic
Streptomyces aureofaciens CMUAc130 and its
antagonism against phytopathogenic fungi
AUTHOR(S): Taechowisan, T.; Peberdy, J. F.; Lumyong, S.
CORPORATE SOURCE: Department of Biology, Faculty of Science, Chiang Mai
University, Chiang Mai, Thailand
SOURCE: Annals of Microbiology (Milano, Italy) (2003), 53(4),
447-461
CODEN: AMNIC7; ISSN: 1590-4261
PUBLISHER: University of Milan, Dep of Food Science and
Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB More than three hundred isolates of endophytic actinomycetes were screened for their potential for chitinase prodn. The strain identified as **Streptomyces aureofaciens** CMUAc130 was the most effective amongst those investigated. This isolate was selected for a more detailed study of chitinase prodn. and its effectiveness in fungal cell wall lysis. Prodn. of the chitinase was optimal with 1% colloidal chitin concn., at 30-40 .degree.C, pH 6.5-7.0 and 100-150 rev min⁻¹ shaking. **N-acetylglucosamine** was a good inducer and expression of the enzyme complex was repressed by several mono- and disaccharides including lactose, mannose, glucose, cellobiose, arabinose, raffinose, sucrose, xylose and fructose. Addn. of pectin, starch and CM-cellulose to the colloidal chitin-contg. medium, increased chitinase prodn. The enzyme tolerated a wide range of temp. (30-50 .degree.C) and pH (5.5-8). Among various divalent cations Hg2+ Cd2+ and Ni2+ completely inhibited the purified enzyme while Mg2+ stimulated its activity. The crude or purified enzyme had potential for cell wall lysis of many phytopathogenic fungi tested.

REFERENCE COUNT: 41 THERE ARE 41 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 9 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1980:194082 HCAPLUS
DOCUMENT NUMBER: 92:194082
TITLE: Induction and its mechanism of a lytic enzyme acting on **Bacillus subtilis** with fungal mycelia
AUTHOR(S): Naka, Hideyuki; Iwamoto, Toru; Inaoka, Megumi
CORPORATE SOURCE: Imabari Meitoku Junior Coll., Imabari, Japan
SOURCE: Hakko Kogaku Kaishi (1980), 58(2), 71-7
CODEN: HKOKDE; ISSN: 0385-6151
DOCUMENT TYPE: Journal
LANGUAGE: Japanese

AB **Streptomyces** Sp. L-13 strain produced a lytic enzyme against *B. subtilis* cells when grown on a medium contg. mycelia of **Aspergillus niger**. Although chitinase and protease were found early in growth, the lytic enzyme and .beta.-1,3-glucanase were produced later. The prodn. of the lytic enzyme and .beta.-1,3-glucanase was enhanced by **N-acetylglucosamine** and the sol. digest from the crude cell wall fraction with protease which was produced from the beginning of growth.

L12 ANSWER 10 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1991:243281 HCAPLUS
DOCUMENT NUMBER: 114:243281
TITLE: Purification and properties of chitinase from **Streptomyces cinereoruber**
AUTHOR(S): Okazaki, Katsuichiro; Tagawa, Kiyoshi
CORPORATE SOURCE: Fac. Agric., Kagawa Univ., Kagawa, 761-07, Japan
SOURCE: Journal of Fermentation and Bioengineering (1991), 71(4), 237-41
CODEN: JFBIEX; ISSN: 0922-338X
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Chitinase (EC 3.2.1.14) was purified from the culture filtrate of *S. cinereoruber*, selected as a microorganism which produces enzymes lysing the cell wall of **Aspergillus niger**, by fractional pptn. with (NH₄)₂SO₄ and column chromatog. on DEAE-cellulose, Sephadex G-100, and CM-Sephadex C-50. The final prepn. was homogeneous in polyacrylamide gel disc electrophoresis. The mol. wt. of the enzyme was .apprx.19,000 and its pI was pH 8.6. The optimum pH and temp. for chitinase activity were 4.5 and at 50.degree., resp. The enzyme was stable at pH 4.0-10.0. The activity was inhibited by Ag⁺, Hg⁺, Hg₂⁺, and p-chloromercuribenzoate. Paper chromatog. anal. demonstrated that the hydrolytic products of colloidal chitin and chitotriose with the enzyme were **N-acetylglucosamine** and chitobiose. The lysis of the *A. niger* cell wall with the enzyme is discussed.

L12 ANSWER 11 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:551280 HCAPLUS
DOCUMENT NUMBER: 139:112733
TITLE: Methods for production of recombinant glycoproteins with mammalian-type carbohydrate structures and their use for production of immunoglobulins
INVENTOR(S): Wildt, Stefan; Miele, Robert Gordon; Nett, Juergen Hermann; Davidson, Robert C.
PATENT ASSIGNEE(S): Glycofi, Inc., USA
SOURCE: PCT Int. Appl., 125 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 8
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2003056914	A1	20030717	WO 2002-US41510	20021224
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,				

LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH,
PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ,
UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY,
KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES,
FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR, BF, BJ,
CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

EP 1467615 A1 20041020 EP 2002-792535 20021224

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, SK

US 2004230042 A1 20041118 US 2003-616082 20030708

PRIORITY APPLN. INFO.:

US 2001-344169P P 20011227
US 2000-214358P P 20000628
US 2000-215638P P 20000630
US 2001-279997P P 20010330
US 2001-892591 A2 20010627
WO 2002-US41510 W 20021224
US 2003-371877 A2 20030220

AB The present invention relates to host cells having modified lipid-linked oligosaccharides which may be modified further by heterologous expression of a set of glycosyltransferases, sugar transporters and mannosidases to become host-strains for the prodn. of mammalian, e.g., human therapeutic glycoproteins. The process provides an engineered host cell which can be used to express and target any desirable gene(s) involved in glycosylation. Host cells with modified lipid-linked oligosaccharides are created or selected. N-glycans made in the engineered host cells have a GlcNAcMan3GlcNAc2 core structure which may then be modified further by heterologous expression of one or more enzymes, e.g., glycosyltransferases, sugar transporters and mannosidases, to yield human-like glycoproteins. For the prodn. of therapeutic proteins, this method may be adapted to engineer cell lines in which any desired glycosylation structure may be obtained. The invention specifically claims use of nucleic acid sequences for gene ALG3 from *Pichia pastoris*. The ALG3 gene encodes the enzyme which transfers a mannose residue to the Man5-GlcNAc2-PP-Dol precursor. The invention also claims use of genetically engineered host cells for recombinant prodn. of Igs. In examples of the invention, a *Pichia pastoris* strain with deletions of genes *alg3* and *och1* was constructed. This strain was transformed with the Kringle 3 domain of human plasminogen as a glycosylation substrate. Mass spectrometric anal. of N-glycans isolated from the kringle 3 glycoproteins showed GlcNAcMan3GlcNAc2 and GlcNAcMan4GlcNAc2 structures which could be further modified in vitro. Addn. of **N-acetylglucosamine** to GlcNAcMan3GlcNAc2 by N-acetylglucosaminyltransferases II and III yields a "bisected" N-glycan, GlcNAc3Man3GlcNAc2, which has been implicated in greater antibody-dependent cellular cytotoxicity. Methods of the invention can be used to engineer a yeast strain capable of producing glycoproteins with bisected N-glycans and expressing Ig mols. with bisected N-glycans attached to asparagine residue 297 in the CH2 portion.

REFERENCE COUNT: 15 THERE ARE 15 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 12 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1971:401545 HCAPLUS

DOCUMENT NUMBER: 75:1545

TITLE: Chemical composition of wild-type and mutant
Aspergillus nidulans cell walls. Nature of
polysaccharide and melanin constituents

AUTHOR(S): Bull, Alan T.

CORPORATE SOURCE: Dep. Agron., Cornell Univ., Ithaca, NY, USA

SOURCE: Journal of General Microbiology (1970), 63(Pt. 1),
75-94

CODEN: JGMIAN; ISSN: 0022-1287

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Chitin and a .beta.-linked glucan were the major chem. components of *A. nidulans* (wild-type strain 13 and an albino mutant) grown in chem. defined media. Enzymic and acid hydrolyses showed galactose, mannose, glucuronic acid, and galactosamine were present. The .beta.-glucan contained (1 .fwdarw. 3) and (1 .fwdarw. 6) linkages and was 2/3 digested by an

exo-.beta.-D-1,3-glucanase prepd. from a cell wall-lysing
Streptomyces. An .alpha.-glucan contg. (1 .fwdarw. 3) linkages
was shown to be a cell-wall component and could be distinguished from
nigeran by ir spectrum and hydrolysis rate. The N-
acetylglucosamine:galactosamine ratio in the A. nidulans cell wall
was 1:32 and the hexosamines were polymer constituents. Protein, lipids,
and in the wild-type, melanin were also cell-wall constituents.

L12 ANSWER 13 OF 37 MEDLINE on STN

ACCESSION NUMBER: 2003395536 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12932365
TITLE: Enhanced enzymatic hydrolysis of langostino shell chitin
with mixtures of enzymes from bacterial and fungal sources.
AUTHOR: Donzelli Bruno G G; Ostroff Gary; Harman Gary E
CORPORATE SOURCE: Department of Horticultural Sciences, Department of Plant
Pathology, Cornell University, Geneva, NY 14456, USA..
bdd1@cornell.edu
SOURCE: Carbohydrate research, (2003 Sep 1) 338 (18) 1823-33.
Journal code: 0043535. ISSN: 0008-6215.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200406
ENTRY DATE: Entered STN: 20030823
Last Updated on STN: 20040625
Entered Medline: 20040624

AB A combination of enzyme preparations from **Trichoderma atroviride**
and **Serratia marcescens** was able to completely degrade high
concentrations (100 g/L) of chitin from langostino crab shells to
N-acetylglucosamine (78%), glucosamine (2%), and
chitobiose (10%). The result was achieved at 32 degrees C in 12 days with
no pre-treatment (size reduction or swelling) of the substrate and without
removal of the inhibitory end-products from the mixture. Enzymatic
degradation of three forms of chitin by **Serratia/**
Trichoderma and **Streptomyces/Trichoderma**
blends was carried out according to a simplex-lattice mixture design.
Fitted polynomial models indicated that there was synergy between
prokaryotic and fungal enzymes for both hydrolysis of crab chitin and
reduction of turbidity of colloidal chitin (primarily endo-type activity).
Prokaryotic/fungal enzymes were not synergistic in degrading chitosan.
Enzymes from prokaryotic sources had much lower activity against chitosan
than enzymes from T. atroviride.

L12 ANSWER 14 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1977:41900 HCAPLUS
DOCUMENT NUMBER: 86:41900
TITLE: An enzymic method for the estimation of the mycelial
content of Shoyu koji and the significance of its
content in Shoyu production
AUTHOR(S): Katou, Takeshi; Kobayashi, Kunio; Izumi, Yasuhiro;
Hanaoka, Yoshio
CORPORATE SOURCE: Res. Lab., Kikkoman Shoyu Co., Ltd., Takasago, Japan
SOURCE: Nippon Nogei Kagaku Kaishi (1976), 50(9), 395-402
CODEN: NNKKAA; ISSN: 0002-1407
DOCUMENT TYPE: Journal
LANGUAGE: Japanese

AB The crude enzyme prepn. contg. .beta.-1,3-glucanase and chitinase, sepd.
from the culture fluid of **Bacillus circulans** (IAM 1165), had
lytic activity on the cell wall of **Aspergillus sojae** X-816 and
liberated **N-acetylglucosamine** [7512-17-6]. By detg.
the liberated **N-acetylglucosamine**, the mycelium
content was calcd. The mycelium content correlated with the proteinase
[9001-92-7] and amylase [9000-92-4] activities, degrdn. rate of total N,
and ratio of amino-N to total N in koji digestion. No relation was obsd.
between spore formation of koji and either mycelial content or the degrdn.
rate of total N.

L12 ANSWER 15 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1972:109677 HCAPLUS
 DOCUMENT NUMBER: 76:109677
 TITLE: Structural studies with the glycopeptides from porcine pancreatic ribonuclease
 AUTHOR(S): Kabasawa, Izumi; Hirs, C. H. W.
 CORPORATE SOURCE: Dep. Biol., Brookhaven Natl. Lab., Upton, NY, USA
 SOURCE: Journal of Biological Chemistry (1972), 247(5), 1610-24
 CODEN: JBCHA3; ISSN: 0021-9258
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Tryptic glycopeptide fractions GP-1, -2, and -3, representative of the heterosaccharides attached to asparagines-21, -34, and -76, resp., in porcine pancreatic RNase were subjected to oxidn. with HIO₄, either alone or in combination with successive, controlled hydrolyses with carefully purified glycosidases. The enzymes used were .alpha.-mannosidase from jack beans, .beta.-mannosidase from the hepatopancreas of Bu-sycotypus [Busycon], .beta.-galactosidase and .beta.-N-acetylglucosaminidase from Diplococcus pneumoniae, .beta.-N-acetylglucosaminidase from *Aspergillus niger*, and neuraminidase from *Vibrio cholerae* [*Vibrio* comma]. The progress of these degradative procedures was monitored by gas-liquid chromatog. of the per-trimethylsilyl derivs. of methylglycosides produced after acid-catalyzed methanolysis. The following were the principal findings and conclusions. 1) Of the 6 mannose residues in GP-2, 4 are .alpha.-linked and 2 are .beta.-linked. These distinct types of residue occur to mutual exclusion in sep. side chains attached to an **N-acetylglucosamine** disaccharide core unit, itself glycosidically linked to asparagine. 2) By the successive action of neuraminidase, .beta.-galactosidase, and .beta.-N-acetylglucosaminidase on GP-1 and GP-3 fractions, peptido-heptasaccharides were obtained which contained identical heterosaccharide moieties composed of 3 residues each of mannose and **N-acetylglucosamine** and 1 residue of fucose. By further degradation of these peptido-heptasaccharides a peptidotet-rasaccharide was obtained with the following partial structure. Fuc(1 .fwdarw. 3,4).beta.GlcNAc(1 .fwdarw. 3).alpha.Man(1 .fwdarw. 3,4)GlcNAc .fwdarw. Asn 3) The heterogeneity of GP-1 and GP-3 fractions may be attributed to variation in the extent to which the common heptasaccharide core moiety in these fractions is substituted with .beta.(1 .fwdarw. 6)-linked **N-acetylglucosamine**-contg. side chains which terminate in sialyl-galactose units. 4) The results suggest that during biosynthesis differentiation of the 2 different types of heterosaccharide side chains in porcine RNase occurs as the 2nd carbohydrate residue is attached to an aspartamido-2-acetamido-1,2-dideoxyglucopy-ranose unit in the peptide chain.

L12 ANSWER 16 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1986:532136 HCAPLUS
 DOCUMENT NUMBER: 105:132136
 TITLE: Production of guanidylfumgin B
 INVENTOR(S): Beppu, Teruhiko; Takesako, Kazutada
 PATENT ASSIGNEE(S): Takara Shuzo Co., Ltd., Japan
 SOURCE: Jpn. Kokai Tokkyo Koho, 6 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 61010595	A2	19860118	JP 1984-130020	19840626
PRIORITY APPLN. INFO.:			JP 1984-130020	19840626

AB The antibiotic guanidylfumgin B (I) is produced by cultivation of **Streptomyces**. Thus, *S. hygroscopicus* was aerobically cultured at 27.degree. for 3 days in a medium contg. starch, glucose, corn steep liquor, yeast ext., **N-acetylglucosamine**, K₂HPO₄, NH₄Cl, FeSO₄, and CaCO₃. The microbial cells were extd. with Me₂CO-H₂O (3:1), and the ext. was worked up by column chromatog. on silica gel with

CHCl₃-MeOH (3:1) and CHCl₃-MeOH-H₂O (5:4:1) to yield I. MIC's of I were Staphylococcus aureus 50, **Bacillus subtilis** 50, **Candida albicans** 100, Trichophyton metagrophytes 6.25, and **Aspergillus oryzae** 25 .mu.g/mL.

L12 ANSWER 17 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2001:767275 HCAPLUS
DOCUMENT NUMBER: 135:287600
TITLE: N-acetylglucosamine enzymic manufacture
INVENTOR(S): Suzuki, Kiyoshi
PATENT ASSIGNEE(S): Seikagaku Kogyo Co., Ltd., Japan
SOURCE: Jpn. Kokai Tokkyo Koho, 7 pp.
CODEN: JKXXAF
DOCUMENT TYPE: Patent
LANGUAGE: Japanese
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2001292791	A2	20011023	JP 2000-112404	20000413
PRIORITY APPLN. INFO.:			JP 2000-112404	20000413

OTHER SOURCE(S): CASREACT 135:287600

AB The N-acetylglucosamine (I) is manufd. with galactose-transferring enzyme from **N-acetylglucosamine** and galactose in the presence of glucose oxidase to remove the glucose, a inhibitor of the reaction, from the reaction system. I is an useful Bifidobacterium growth promoter. Prepn. of I with .beta.-galactosidase of **Bacillus circulans** was shown.

L12 ANSWER 18 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1980:462284 HCAPLUS
DOCUMENT NUMBER: 93:62284
TITLE: Production, biosynthesis and mode of action of **Bacillus** oligopeptide antibiotics. 9
AUTHOR(S): Vandamme, E. J.
CORPORATE SOURCE: Lab. Alg. Ind. Microbiol., Rijksuniv. Gent, Ghent, Belg.
SOURCE: Revue des Fermentations et des Industries Alimentaires (1979), 34(6), 167-73
CODEN: RFIAAQ; ISSN: 0035-2071
DOCUMENT TYPE: Journal
LANGUAGE: Dutch

AB Bacilysin (I) [29393-20-2] showed much greater activity in vitro than anticapsin [28978-07-6] against **Candida albicans** and various bacteria such as Staphylococcus aureus, Escherichia coli, and Salmonella. Resistance to I occurred frequently in these organisms, except for E. coli. Growing cultures of I-sensitive strains of Staphylococcus aureus, that are not resistant strains, slowly inactivated I by converting it to anticapsin through cleavage and L-alanine residue. This conversion was also performed by cell-free exts. both sensitive and resistant strains, and was catalyzed by a metal ion-requiring dipeptidase [74315-97-2]. The antibacterial action of I against Staphylococcus aureus was antagonized by various dipeptides and amino acids (which may compete with I for a peptide uptake system) and by D-(+)-glucosamine-HCl [66-84-2] and **N-acetylglucosamine** [7512-17-6]. The antibacterial activity of I may be related to an effect on D-glucosamine biosynthesis. D-Glucosamine-6-phosphate synthetase [9030-45-9] was demonstrated in Staphylococcus aureus.

L12 ANSWER 19 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2002:420648 HCAPLUS
DOCUMENT NUMBER: 137:136845
TITLE: Comparative study of the reaction mechanism of family 18 chitinases from plants and microbes
AUTHOR(S): Sasaki, Chiye; Yokoyama, Ai; Itoh, Yoshifumi; Hashimoto, Masayuki; Watanabe, Takeshi; Fukamizo, Tamo
CORPORATE SOURCE: Laboratory of Enzyme System Science, Department of Food and Nutrition, Kinki University, Nara, 631-8505,

SOURCE: Japan
Journal of Biochemistry (Tokyo, Japan) (2002), 131(4),
557-564
CODEN: JOBIAO; ISSN: 0021-924X
PUBLISHER: Japanese Biochemical Society
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Hydrolytic mechanisms of family 18 chitinases from rice (*Oryza sativa* L.) and *Bacillus* circulans WL-12 were comparatively studied by a combination of HPLC anal. of the reaction products and theor. calcn. of reaction time-courses. All of the enzymes tested produced .beta.-anomers from chitin hexasaccharide [(GlcNAc)₆], indicating that they catalyze the hydrolysis through a retaining mechanism. The rice chitinases hydrolyzed predominantly the fourth and fifth glycosidic linkages from the nonreducing end of (GlcNAc)₆, whereas *B. circulans* chitinase A1 hydrolyzed the second linkage from the nonreducing end. In addn., the *Bacillus* enzyme efficiently catalyzed transglycosylation, producing significant amts. of chitin oligomers larger than the initial substrate, but the rice chitinases did not. The time-courses of (GlcNAc)₆ degrdn. obtained by HPLC were analyzed by theor. calcn., and the subsite structures of the rice chitinases were identified to be (-4)(-3)(-2)(-1)(+1)(+2). From the HPLC profile of the reaction products previously reported, family 18 chitinase from rubber tree (*Hevea brasiliensis*) was estd. to have the same type of subsite structure. Theor. anal. of the reaction time-course for the *Bacillus* enzyme revealed that the enzyme has (-2)(-1)(+1)(+2)(+3)(+4)-type subsite structure, which is identical to that of fungal chitinase from *Coccidioides immitis*. The *Bacillus* enzyme also resembled the fungal chitinase in its transglycosylation activity. Minor structural differences between plant and microbial enzymes appear to result in such functional variations, even though all of these chitinases are classified into the identical family of glycosyl hydrolases.

REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 20 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1996:308497 HCAPLUS

DOCUMENT NUMBER: 124:351595

TITLE: Biotreatment of marine crustacean and chicken egg shell waste

AUTHOR(S): Healy, M. G.; Bustos, R. O.; Solomon, S. E.; Devine, C.; Healy, A.

CORPORATE SOURCE: Department Chemical Engineering, Queen's University Belfast, UK

SOURCE: Environmental Biotechnology: Principles and Applications, [Papers presented at the International Symposium on Environmental Biotechnology], Waterloo, Ont., July 4-8, 1994 (1996), Meeting Date 1994, 302-319. Editor(s): Moo-Young, Murray; Anderson, William A.; Chakrabarty, Ananda M. Kluwer: Dordrecht, Neth.
CODEN: 62UGA4

DOCUMENT TYPE: Conference

LANGUAGE: English

AB A microbial/enzymic approach to biol. deproteinize shell waste from the marine crustacean, *Nephrops norvegicus*, and shell waste from chicken (*Gallus domesticus*) eggs, was studied with particular emphasis on methods to produce chitin from prawn shells. Com. available silage inoculants (mixed bacterial and bacterial/fungi cultures) and proteolytic strains of *Pseudomonas* (*Xanthomonas*) maltophilia were evaluated for their potential to deproteinize acid demineralized prawn shell waste. Highest deproteinization of prawn shell was achieved using *Bacillus subtilis* strain HB-3B and a mixed culture inoculant, BAFP-202, while *Pseudomonas* maltophilia strain 9204 best deproteinized chicken shell waste.

L12 ANSWER 21 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1991:205551 HCAPLUS

DOCUMENT NUMBER: 114:205551

TITLE: Galactooligosaccharide conjugates with sugars,
alcohols and nucleosides prepared with microorganisms
INVENTOR(S): Oonishi, Norimase; Yokozeki, Kenzo
PATENT ASSIGNEE(S): Ajinomoto Co., Inc., Japan
SOURCE: Fr. Demande, 19 pp.
CODEN: FRXXBL
DOCUMENT TYPE: Patent
LANGUAGE: French
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
FR 2640997	A1	19900629	FR 1989-17127	19891222
FR 2640997	B1	19950421		
JP 02072890	A2	19900313	JP 1988-324855	19881222
JP 03035787	A2	19910215	JP 1989-168103	19890629
PRIORITY APPLN. INFO.:			JP 1988-324855	A 19881222
			JP 1989-168103	A 19890629
			JP 1988-118160	A1 19880517

AB Conjugates of oligogalactosides ((Gal)n-R; n = 1-4, R = sugar, sugar alc., alc., nucleoside) are prepd. from a galactose donor such as o-nitrophenyl galactoside and an appropriate acceptor using a microorganism such as Rhodotorula, Cryptococcus, Corynebacterium and Sirobasidium. Cells of R. minuta, Stergimatomyces elviae and Sirobasidium magnum grown in a yeast ext./peptone/salts medium with galactose and glycerol as C sources were resuspended in phosphate buffer contg. lactose 22 g/dL and incubated at 50.degree. for 2 days. Approx. half of the lactose was metabolized (recovery 9.6-11.2 g/dL) with recoveries of galactose trisaccharide of 8-9.3 g/dL and of the tetrasaccharide of 0-1.7 g/dL. When R. minuta was incubated in a medium contg. 2.5 g lactose and 5 g inosine/50 mL resulted in the formation of the mono-, di-, and .gtoreq.3 galactosides 1.4, 0.8 and 0.4 g/dL resp.

L12 ANSWER 22 OF 37 MEDLINE on STN

ACCESSION NUMBER: 2002736587 MEDLINE

DOCUMENT NUMBER: PubMed ID: 12499170

TITLE: Mannopeptimycins, new cyclic glycopeptide antibiotics produced by **Streptomyces** hygrosopicus LL-AC98: antibacterial and mechanistic activities.

AUTHOR: Singh M P; Petersen P J; Weiss W J; Janso J E; Luckman S W; Lenoy E B; Bradford P A; Testa R T; Greenstein M

CORPORATE SOURCE: Natural Products Microbiology. Antibacterial Research, Infectious Disease Section, Wyeth Research, Pearl River, New York 10965, USA.. singhm@wyeth.com

SOURCE: Antimicrobial agents and chemotherapy, (2003 Jan) 47 (1) 62-9.

Journal code: 0315061. ISSN: 0066-4804.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200306

ENTRY DATE: Entered STN: 20021227

Last Updated on STN: 20030604

Entered Medline: 20030603

AB Mannopeptimycins alpha, beta, gamma, delta, and epsilon are new cyclic glycopeptide antibiotics produced by **Streptomyces** hygrosopicus LL-AC98. Mannopeptimycins gamma, delta, and epsilon, which have an isovaleryl substitution at various positions on the terminal mannose of the disaccharide moiety, demonstrated moderate to good antibacterial activities. Mannopeptimycin epsilon was the most active component against methicillin-resistant staphylococci and vancomycin-resistant enterococci (MICs, 2 to 4 micro g/ml for staphylococci and streptococci and 4 to 32 micro g/ml for enterococci), while mannopeptimycins gamma and delta were two- to fourfold less active. Mannopeptimycins alpha and beta, which lack the isovaleryl substitution and the disaccharide moiety, respectively, had poor antibacterial activities. The in vivo efficacies of the mannopeptimycins in Staphylococcus aureus mouse protection studies

paralleled their in vitro activities. The median effective doses of mannopeptimycins gamma, delta, and epsilon were 3.8, 2.6, and 0.59 mg/kg of body weight, respectively. The mannopeptimycins were inactive against cell wall-deficient *S. aureus* and caused spheroplasting of *Escherichia coli* imp similar to that observed with penicillin G in an osmotically protective medium. Mannopeptimycin delta rapidly inhibited [(3)H]

N-acetylglucosamine incorporation into peptidoglycan in *Bacillus subtilis* and had no effect on DNA, RNA, or protein biosynthesis. On the basis of the observations presented above, an effect on cell wall biosynthesis was suggested as the primary mode of action for mannopeptimycin delta. The mannopeptimycins were inactive against *Candida albicans*, did not initiate hemolysis of human erythrocytes, and did not promote potassium ion leakage from *E. coli* imp, suggesting a lack of membrane damage to prokaryotic or eukaryotic cells.

L12 ANSWER 23 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1996:329512 HCAPLUS

DOCUMENT NUMBER: 125:5106

TITLE: The nikkomycin story

AUTHOR(S): Fiedler, Hans-Peter

CORPORATE SOURCE: Biologisches Institut, Universitat Tübingen, Tübingen, D-72076, Germany

SOURCE: Sekundaermetabolismus bei Mikroorganismen: Beiträege zur Forschung, [Proceedings of the International Conference on Microbial Secondary Metabolism], Interlaken, Switz., Oct. 1994 (1995), Meeting Date 1994, 79-89. Editor(s): Kuhn, Willi; Fiedler, Hans-Peter. Attempto Verlag: Tübingen, Germany. CODEN: 62XFAG

DOCUMENT TYPE: Conference; General Review

LANGUAGE: English

AB A review with 51 refs. Nikkomycins are produced by *Streptomyces tendae* strain TU 901. They belong to the group of nucleoside-peptide antibiotics and are similar in structure to polyoxins. The site of action involves the biosynthesis of chitin in the cell wall because of a structural similarity to UDP-**N-acetylglucosamine**. For this reason, nikkomycins show no biol. activity against Gram-pos. and Gram-neg. bacteria, but are distinguished by high antifungal, insecticidal and acaricidal activities. No toxicity against plants, fish and mammals are obsd. In combination with other inhibitors of fungal cell wall biosynthesis, e.g. azoles, papulacandin B and cilofungin, an excellent synergistic action was obsd. against *Candida albicans*.

L12 ANSWER 24 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2002:481348 HCAPLUS

DOCUMENT NUMBER: 137:155129

TITLE: Use of Ionic Liquids to Increase the Yield and Enzyme Stability in the .beta.-Galactosidase Catalyzed Synthesis of N-Acetylglucosamine

AUTHOR(S): Kaftzik, Nicole; Wasserscheid, Peter; Kragl, Udo

CORPORATE SOURCE: Department of Chemistry, Rostock University, Rostock, 18051, Germany

SOURCE: Organic Process Research & Development (2002), 6(4), 553-557

CODEN: OPRDFK; ISSN: 1083-6160

PUBLISHER: American Chemical Society

DOCUMENT TYPE: Journal

LANGUAGE: English

OTHER SOURCE(S): CASREACT 137:155129

AB The use of ionic liqs. as alternative solvents for enzyme catalysis was investigated. .beta.-Galactosidase from *Bacillus circulans* catalyzes the synthesis of N-acetylglucosamine starting from lactose and **N-acetylglucosamine** in a transglycosylation reaction. The addn. of 25% vol./vol. of 1,3-di-methyl-imidazol-Me sulfate [MMIM] [MeSO₄] as a water-miscible ionic liq. suppresses the secondary hydrolysis of the formed product, resulting in doubling the yield to almost 60%. The enzyme can be reused several times after ultrafiltration of the reaction mixt. without loss of activity. Results of different amts. of ionic liqs. in the reaction medium on the thermostability of the galactosidase as well

as on oxidoreductases are presented as well.

REFERENCE COUNT: 53 THERE ARE 53 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L12 ANSWER 25 OF 37 MEDLINE on STN

ACCESSION NUMBER: 2004575023 IN-PROCESS

DOCUMENT NUMBER: PubMed ID: 15502369

TITLE: Purification and antifungal activity of recombinant
chitinase from *Escherichia coli* carrying the family 19
chitinase gene of **Streptomyces** sp. J-13-3.

AUTHOR: Yamashita Yousuke; Okazaki Katsuichiro

CORPORATE SOURCE: Department of Life Sciences, Faculty of Agriculture, Kagawa
University, Miki, Kagawa 761-0795, Japan.

SOURCE: Bioscience, biotechnology, and biochemistry, (2004 Oct) 68
(10) 2193-6.

Journal code: 9205717. ISSN: 0916-8451.

PUB. COUNTRY: Japan

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: NONMEDLINE; IN-PROCESS; NONINDEXED; Priority Journals

ENTRY DATE: Entered STN: 20041123

Last Updated on STN: 20041219

AB A recombinant chitinase was purified from the cell extract of *Escherichia coli* JM109 transformed by plasmid pUC19 carrying the gene encoding family 19 chitinase of **Streptomyces** sp. J-13-3 by column chromatography on DEAE-Sepharose, CM-Sepharose, and Bio-Gel P-100. The final preparation was homogenous in polyacrylamide gel electrophoresis. The molecular weight of the purified enzyme was estimated to be 32,000. The recombinant chitinase hydrolyzed the trimer to hexamer of **N-acetylglucosamine** and had the identical N-terminal amino acid sequence of the mature protein, indicating removal of the signal sequence by *E. coli* signal peptidase. The fungal growth in well (200 microl of medium) of microplate by measurement of absorbance at 595 nm indicated that the chitinase (10 microg) completely and half inhibited growth of **Trichoderma reesei** and **Aspergillus niger** respectively.

L12 ANSWER 26 OF 37 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 2000-05519 BIOTECHDS

TITLE: Purification and characterization of two chitinases from
Streptomyces albobovineus S-22;
with the potential for cell wall lysis of fungal
pathogenesis

AUTHOR: El-Sayed E S A; Ezzat S M; Ghaly M F; Mansour M; El-Bohey M A

CORPORATE SOURCE: Univ.Zagazig

LOCATION: Botany Department, Faculty of Science, Zagazig University,
Zagazig, Egypt.

SOURCE: World J.Microbiol.Biotechnol.; (2000) 16, 1, 87-89

CODEN: WJMBEY

ISSN: 0959-3993

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Two chitinases (EC-3.2.1.14), A and B, were purified from the culture supernatant of **Streptomyces albobovineus** S-22 by ammonium sulfate fraction (80%) and Sephadex G-200 gel filtration. Both enzymes had molecular weights estimated to be 43,000 and 45,000 by SDS-PAGE. The enzymes were active at 40 deg and pH 5.6 after 120 min, and stable at temperatures below 40 deg in the absence of chitin. The purified enzyme had potential for cell wall lysis of fungal pathogenesis tested. The data in this work indicated that the relative activities of purified chitinase of *S. albobovineus* S-22 significantly increased with increasing incubation times at 40 deg and their maximal value reached 100% at 120 min, then slightly decreased as the times increased. This could be due to denaturation of the enzyme protein. The highest amounts of **N-acetylglucosamine** were determined from the hydrolysis of **Aspergillus niger** mycelia followed by *Alternaria* sp., *Fusarium oxysporum*, *Fusarium lycopersici* then **Trichoderma** sp. and finally **Aspergillus flavus**. This indicated the chitinases of *S. albobovineus* S-22 may be used for biocontrol of phytogetic fungi. (6 ref)

L12 ANSWER 27 OF 37 MEDLINE on STN

ACCESSION NUMBER: 2004090764 MEDLINE

DOCUMENT NUMBER: PubMed ID: 14981297

TITLE: Molecular cloning and expression of the gene encoding family 19 chitinase from **Streptomyces** sp. J-13-3.

AUTHOR: Okazaki Katsuichiro; Yamashita Yousuke; Noda Minoru; Sueyoshi Noriyuki; Kameshita Isamu; Hayakawa Shigeru

CORPORATE SOURCE: Department of Life Sciences, Faculty of Agriculture, Kagawa University, Japan.. okazaki@ag.kagawa-u.ac.jp

SOURCE: Bioscience, biotechnology, and biochemistry, (2004 Feb) 68 (2) 341-51.

Journal code: 9205717. ISSN: 0916-8451.

PUB. COUNTRY: Japan

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

OTHER SOURCE: GENBANK-AB116547

ENTRY MONTH: 200410

ENTRY DATE: Entered STN: 20040225

Last Updated on STN: 20041026

Entered Medline: 20041025

AB The gene encoding chitinase from **Streptomyces** sp. (strain J-13-3) was cloned and its nucleotide structure was analyzed. The chitinase consisted of 298 amino acids containing a signal peptides (29 amino acids) and a mature protein (269 amino acids), and had calculated molecular mass of 31,081 Da. The calculated molecular mass (28,229 Da) of the mature protein was almost same as that of the native chitinase determined by matrix-assisted laser desorption ionization time-of-flight mass spectrometer. Comparison of the encoded amino acid sequences with those of other chitinases showed that J-13-3 chitinase was a member of the glycosyl-hydrolase family 19 chitinases and the mature protein had a chitin binding domain (65 amino acids) containing AKWWTQ motif and a catalytic domain (204 amino acids). The J-13-3 strain had a single chitinase gene. The chitinase (298 amino acids) with C-terminal His tag was overexpressed in *Escherichia coli* BL21(DE3) cells. The recombinant chitinase purified from the cell extract had identical N-terminal amino acid sequence of the mature protein in spite of confirmation of the nucleotide sequence, suggesting that the signal peptide sequence is successfully cut off at the predicted site by signal peptidase from *E. coli* and will be a useful genetic tool in protein engineering for production of soluble recombinant protein. The optimum temperature and pH ranges of the purified chitinase were at 35-40 degrees C and 5.5-6.0, respectively. The purified chitinase hydrolyzed colloidal chitin and trimer to hexamer of **N-acetylglucosamine** and also inhibited the hyphal extension of *Tricoderma reesei*.

L12 ANSWER 28 OF 37 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 2001-10302 BIOTECHDS

TITLE: Enzymes of **Candida albicans** cell-wall lytic system produced by **Streptomyces thermodiastaticus**; selection of culture medium and conditions for lytic enzyme production

AUTHOR: Mansour F A; *Mohamedin A H

CORPORATE SOURCE: Univ.Mansoura

LOCATION: Department of Botany, Faculty of Science, Mansoura University, Mansoura, Egypt..

SOURCE: Acta Microbiol.Immunol.Hung.; (2001) 48, 1, 53-65

CODEN: 0165P

ISSN: 1217-8950

DOCUMENT TYPE: Journal

LANGUAGE: English

AB 24 Thermophilic actinomycetes strains were isolated from Egyptian soil samples and screened for the ability to degrade cell walls of **Candida albicans**. A selected isolate was taxonomically characterized as belonging to **Streptomyces thermodiastaticus**. The effects of incubation period, culture medium pH, incubation temp., different C-sources (C. albicans cell wall, glucose, starch, chitin, mannan, cellobiose, CM-cellulose, sucrose, fructose, glycerol, maltose,

N-acetylglucosamine, mannitol), different N-sources (KNO₃, NaNO₃, casein, (NH₄)₂SO₄, NH₄NO₃, NH₄Cl, (NH₄)₃PO₄, urea and peptone), different P-sources (NaH₂PO₄, Na₂HPO₄, Na₃PO₄, KH₂PO₄, K₂HPO₄, K₃PO₄, NH₄H₂PO₄, (NH₄)₂HPO₄ and (NH₄)₃PO₄, different microelements, Tween-80, yeast extract, methionine and casamino acids were examined. Highest lytic activity was obtained when *S. thermophilus* was grown in a medium consisting of 1% *C. albicans* cell wall, 0.1% NaNO₃, 0.1% KH₂PO₄, 0.0001% ZnSO₄ and 0.1% Tween-80, the pH of the medium was adjusted to 5.5, and cultures were incubated at 50 deg for 18 hr. The lytic system produced by *S. thermophilus* contained chitinolytic and proteolytic activities. (39 ref)

L12 ANSWER 29 OF 37 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1996:485508 HCAPLUS
DOCUMENT NUMBER: 125:273645
TITLE: Syntheses of oligosaccharide components of glycoconjugates using glycosidases
AUTHOR(S): Fujimoto, Hiroshi; Isomura, Megumi; Ajisaka, Katsumi
CORPORATE SOURCE: Meiji Inst. Health Sci., Meiji Milk Prod. Co., Ltd., Odawara, 250, Japan
SOURCE: Oyo Toshitsu Kagaku (1996), 43(2), 265-272
CODEN: OTKAE3; ISSN: 1340-3494
PUBLISHER: Nippon Oyo Toshitsu Kagakkai
DOCUMENT TYPE: Journal; General Review
LANGUAGE: Japanese

AB A review with 18 refs. Enzymic synthesis of oligosaccharide components of glycoconjugates was studied by using 2 types of reactions of glycosidases: reverse hydrolysis reactions and transglycosylation. Mannooligosaccharides were synthesized by incubating mannose(Man) with .alpha.-mannosidase from **Aspergillus niger** at 50.degree. and products were sepd. by active-C column chromatog. followed by HPLC. Following products were obtained from 10 g Man: Man.alpha.1-2Man 290 mg, Man.alpha. 1-2Man.alpha. 1-2Man 14 mg, Man.alpha. 1-6Man 2.2 g, Man.alpha. 1-3Man 33 mg, and Man.alpha. 1-2Man .alpha. 1-6Man 3 mg. The action of .beta.-N-acetylglucosaminidase from jack bean on a concd. mixt. of **N-acetylglucosamine** (GlcNAc) and Man gave GlcNAc.beta. 1-2Man (I) and GlcNAc.beta. 1-6Man (II) in yields .apprx.0.2 and 1.7%, resp. The regiospecific introduction of galactose (Gal) residue from p-nitrophenyl .beta.-galactoside (III) to nonreducing ends of I and II were carried out by the transglycosylation of .beta.-galactosidase from **Bacillus circulans** to give Gal.beta. 1-4GlcNAc.beta. 1-2Man and Gal1-4GlcNAc.beta.1-6Man. Gal.beta.1-4GlcNAc was synthesized through transgalactosylation from III by *Diplococcus pneumoniae* .beta.-galactosidase, and sialyl residue was introduced to the product from sialic acid dimer [(Neu5Ac)₂] by transfer action by sialidase from Newcastle disease virus, to give Neu5Ac.alpha.2-3Gal.beta.1-4GlcNAc. The transfer of fucose (Fuc) residue from p-nitrophenyl .alpha.-fucoside to Glc, GlcNA, and other acceptors was tested by .alpha.-fucosidase of several origins, and Fuc.alpha.1-3Glc and Fuc.alpha.1-3GlcNAc were obtained in high yields (61 and 58%, resp.) by the enzyme from **Aspergillus niger**. These oligosaccharides can be used for chem. synthesis of glycoconjugates.

L12 ANSWER 30 OF 37 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1998:671529 SCISEARCH
THE GENUINE ARTICLE: 114PZ
TITLE: Chitinolytic activity in *Chromobacterium violaceum*: Substrate analysis and regulation by quorum sensing
AUTHOR: Chernin L S (Reprint); Winson M K; Thompson J M; Haran S; Bycroft B W; Chet I; Williams P; Stewart G S A B
CORPORATE SOURCE: HEBREW UNIV JERUSALEM, FAC AGR FOOD & ENVIRONM QUAL SCI, OTTO WARBURG CTR BIOTECHNOL AGR, POB 12, IL-76100 REHOVOT, ISRAEL (Reprint); HEBREW UNIV JERUSALEM, FAC AGR FOOD & ENVIRONM QUAL SCI, DEPT PLANT PATHOL & MICROBIOL, IL-76100 REHOVOT, ISRAEL; UNIV NOTTINGHAM, SCH BIOL, LOUGHBOROUGH LE12 5RD, LEICS, ENGLAND; UNIV NOTTINGHAM, SCH PHARMACEUT SCI, NOTTINGHAM NG7 2RD, ENGLAND
COUNTRY OF AUTHOR: ISRAEL; ENGLAND

SOURCE: JOURNAL OF BACTERIOLOGY, (SEP 1998) Vol. 180, No. 17, pp. 4435-4441.
Publisher: AMER SOC MICROBIOLOGY, 1325 MASSACHUSETTS AVENUE, NW, WASHINGTON, DC 20005-4171.
ISSN: 0021-9193.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 60

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Quorum sensing control mediated by N-acyl homoserine lactone (AHL) signaling molecules has been established as a key feature of the regulation of exoenzyme production in many gram-negative bacteria. In *Chromobacterium violaceum* ATCC 31532 a number of phenotypic characteristics, including production of the purple pigment violacein, hydrogen cyanide, antibiotics, and exoproteases are known to be regulated by the endogenous AHL N-hexanoyl-L-homoserine lactone (HHL). In this study we show that *C. violaceum* produces a set of chitinolytic enzymes whose production is regulated by HHL. The chitinolytic activity was induced in strains grown in the presence of chitin as the sole carbon source and quantitated in the secreted proteins by using p-nitrophenol analogs of disaccharide, trisaccharide, and tetrasaccharide oligomers of **N-acetylglucosamine**. By using 4-methylumbelliferyl analogs of the same oligomers of **N-acetylglucosamine** as substrates for proteins separated and renatured by sodium dodecyl sulfate-polyacrylamide gel electrophoresis, at least six enzymes were detected: a chitinase with high specificity to a dimeric substrate of 87 kDa, two N-acetylglucosaminidases with apparent molecular masses of 162 and 133 kDa, two endochitinases of 108 and 67 kDa, and a chitobiosidase of 56 kDa. In addition, two unidentified bands of >205 kDa were found where a tetrameric chitin derivative was used as a substrate. A pleiotropic mini-Tn5 mutant of *C. violaceum* (CV026) that is defective in HHL production and other quorum-sensing-regulated factors was also found to be completely deficient in chitinolytic activity. Growth of this mutant on minimal medium with chitin supplemented with culture supernatant from the *C. violaceum* wild-type strain or 10 μ M synthetic HHL restored chitinase production to the level shown by the parental strain. These results constitute the most complete evidence so far for regulation of chitinolytic activity by AHL signaling in a gram-negative bacterium.

L12 ANSWER 31 OF 37 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 1997-12390 BIOTECHDS

TITLE: Production of N-acetyl-D-glucosamine;
by chitin hydrolysis with chitinase, beta-N-acetylhexosaminidase or beta-N-acetyl-D-glucosaminidase in a sequential two-stage packed bed and stirred tank fermentor

AUTHOR: Haynes C A; Aloise P; Creagh A L
PATENT ASSIGNEE: Univ.British-Columbia
LOCATION: Vancouver, British Columbia, Canada.
PATENT INFO: WO 9731121 28 Aug 1997
APPLICATION INFO: WO 1997-CA115 20 Feb 1997
PRIORITY INFO: US 1996-603360 20 Feb 1996
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1997-448367 [41]

AB A method for the production of N-acetyl-D-glucosamine is claimed, which involves enzymatically hydrolyzing chitin with an ensemble of chitinolytic enzymes obtained by fermenting chitin obtained from the exoskeletons of crustacea with a selected organism to induce the production of chitinase (EC-3.2.1.14) and beta-N-acetylhexosaminidase (EC-3.2.1.52) or beta-N-acetyl-D-glucosaminidase (EC-3.2.1.30), and isolating the produced N-acetyl-D-glucosamine. Also claimed are: (a) a method for the production of the chitinolytic enzymes, which involves introducing a chitin-containing substance into a fermentor, inoculating with a prokaryote (preferably *Serratia marcescens*, *Streptomyces lividans* or *Enterobacter liquefaciens*) or a eukaryote (preferably *Trichoderma harzianum* or *Myrothecium verrucaria*) and recovering the enzymes from the culture; and (b) a

chitin-containing solid substrate, a defined carbohydrate-free medium and an organism which produces chitin-degrading and chitobiose-degrading enzymes in a sequential two-stage fermentor, preferably comprising of a packed bed fermentor and a stirred tank fermentor. (82pp)

L12 ANSWER 32 OF 37 MEDLINE on STN

ACCESSION NUMBER: 76216534 MEDLINE

DOCUMENT NUMBER: PubMed ID: 819623

TITLE: Antimicrobial activities and antagonists of bacilysin and anticapsin.

AUTHOR: Kenig M; Abraham E P

SOURCE: Journal of general microbiology, (1976 May) 94 (1) 37-45.
Journal code: 0375371. ISSN: 0022-1287.

PUB. COUNTRY: ENGLAND: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 197608

ENTRY DATE: Entered STN: 19900313

Last Updated on STN: 19900313

Entered Medline: 19760823

AB The dipeptide antibiotic bacilysin is active against a wide range of bacteria and against **Candida albicans**. Its C-terminal amino acid, anticapsin, is a very poor antibacterial agent. The activities of both substances are strongly dependent on the nature of the culture medium. In a minimal medium the minimum inhibitory concentration for bacilysin with *E. coli* B is 10(-3) mug ml(-1). The action of bacilysin amino acids. With several bacteria, bacilysin-resistant mutants are found in unusually large numbers. It is suggested that peptide and amino acid transport systems play a role in these phenomena. The antimicrobial action of bacilysin is also inhibited by glucosamine and **N-acetylglucosamine**. This antibiotic may therefore interfere with glucosamine synthesis and thus with the synthesis of microbial cell walls.

L12 ANSWER 33 OF 37 MEDLINE on STN

ACCESSION NUMBER: 91012496 MEDLINE

DOCUMENT NUMBER: PubMed ID: 2120441

TITLE: Antimicrobial properties of N3-(iodoacetyl)-L-2,3-diaminopropanoic acid-peptide conjugates.

AUTHOR: Andruszkiewicz R; Chmara H; Milewski S; Zieniawa T; Borowski E

CORPORATE SOURCE: Department of Pharmaceutical Technology and Biochemistry, Technical University of Gdansk, Poland.

SOURCE: Journal of medicinal chemistry, (1990 Oct) 33 (10) 2755-9.
Journal code: 9716531. ISSN: 0022-2623.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199011

ENTRY DATE: Entered STN: 19910117

Last Updated on STN: 19980206

Entered Medline: 19901120

AB Six peptide conjugates consisting of either norvaline, methionine, or lysine and N3-(iodoacetyl)-L-2,3-diaminopropanoic acid--a strong, irreversible inactivator of bacterial and fungal glucosamine-6-phosphate synthase--were synthesized and their antibacterial and antifungal activities were evaluated. Antimicrobial potencies of these peptides were correlated with their transport and cleavage rates inside the cells. Bacteriolysis of *Bacillus pumilus* cells and inhibition of [14C]glucose incorporation into cell-wall polysaccharides of *Candida albicans* as a result of glucosamine 6-phosphate inactivation were also observed. Reversal of growth inhibitory effect of these peptides by **N-acetylglucosamine** in bacteria and fungi suggests the effective delivery of N3-iodoacetyl-L-2,3-diaminopropanoic acid into the cell by a peptide-transport system.

L12 ANSWER 34 OF 37 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1998:472473 SCISEARCH
THE GENUINE ARTICLE: ZU399
TITLE: The molecular biology of chitin digestion
AUTHOR: CohenKupiec R (Reprint); Chet I
CORPORATE SOURCE: HEBREW UNIV JERUSALEM, FAC AGR, OTTO WARBURG CTR AGR
BIOTECHNOL, POB 12, IL-76100 REHOVOT, ISRAEL (Reprint)
COUNTRY OF AUTHOR: ISRAEL
SOURCE: CURRENT OPINION IN BIOTECHNOLOGY, (JUN 1998) Vol. 9, No.
3, pp. 270-277.
Publisher: CURRENT BIOLOGY LTD, 34-42 CLEVELAND STREET,
LONDON W1P 6LB, ENGLAND.
ISSN: 0958-1669.
DOCUMENT TYPE: General Review; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 52

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Chitinases catalyze the hydrolysis of chitin, an unbranched polymer of
beta-1,4-**N-acetylglucosamine**. In recent years,
soil-borne microorganisms that produce chitinases are considered as
potential biocontrol agents against fungi and nematodes which cause
diseases of agricultural crops. Chitinases also play an important
physiological and ecological role in ecosystems as recyclers of chitin, by
generating carbon and nitrogen sources. Many chitinases of varied
organisms have been isolated and their corresponding genes cloned.

L12 ANSWER 35 OF 37 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 1991-04749 BIOTECHDS
TITLE: Promotive and inhibitory effects of raw starch adsorbable
fragments from pancreatic alpha-amylase on enzymatic
digestions of raw starch;
Gp-pan-I and Gp-pan-P production
AUTHOR: Hayashida S; Teramoto Y; Kira I
LOCATION: Department of Agricultural Chemistry, Kyushu University,
Kukuoka 812, Japan.
SOURCE: Agric.Biol.Chem.; (1991) 55, 1, 1-6
CODEN: ABCHA6
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The purification of 2 enzymatically inactive fragments, designated as
Gp-pan-P and Gp-pan-I, derived from tryptic-digested pig pancreatic
alpha-amylase (EC-3.2.1.1) and their functions are described. These 2
glycopeptide fragments were purified with Sephadex G-75, DEAE-Sephadex
A-50, and HPLC. The mol.wt. of Gp-pan-P and Gp-pan-I were determined by
SDS-PAGE to be 20,000 and 30,000, respectively. The carbohydrate
contents of Gp-pan-P and Gp-pan-I were 10 and 7%, respectively. These
carbohydrates were composed of **N-acetylglucosamine**,
fucose, galactose, and mannose. Neither Gp-pan-P or Gp-pan-I had
alpha-amylase activity on their own but they were specifically adsorbed
onto raw starch. Gp-pan-P promoted raw starch digestion of
Aspergillus awamori var. kawachi glucoamylase-I (EC-3.2.1.3) and
Bacillus subtilis 65 alpha-amylase. Gp-pan-I inhibited the
digestion of raw maize (*Zea mays*) starch by these enzymes. The enzyme
fragments had N-terminal amino acid Gly-Trp and Ala-Val, and C-terminal
amino acids Gly-Arg and Ile-Lys. (23 ref)

L12 ANSWER 36 OF 37 EMBASE COPYRIGHT 2005 ELSEVIER INC. ALL RIGHTS RESERVED.
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ACCESSION NUMBER: 82019930 EMBASE
DOCUMENT NUMBER: 1982019930
TITLE: Deflectins, new antimicrobial azaphilones from *Aspergillus*
deflectus.
AUTHOR: Anke H.; Kemmer T.; Hoefle G.
CORPORATE SOURCE: Inst. Biol. II, Univ. Tübingen D-7400, Germany
SOURCE: Journal of Antibiotics, (1981) 34/8 (923-928).
CODEN: JANTAJ
COUNTRY: Japan
DOCUMENT TYPE: Journal
FILE SEGMENT: 037 Drug Literature Index

004 Microbiology
030 Pharmacology
LANGUAGE: English

L12 ANSWER 37 OF 37 EMBASE COPYRIGHT 2005 ELSEVIER INC. ALL RIGHTS RESERVED.
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ACCESSION NUMBER: 88020894 EMBASE
DOCUMENT NUMBER: 1988020894
TITLE: Synthesis and biological properties of N3-(4-methoxyfumaryl)-L-2,3-diaminopropanoic acid dipeptides, a novel group of antimicrobial agents.
AUTHOR: Andruszkiewicz R.; Chmara H.; Milewski S.; Borowski E.
CORPORATE SOURCE: Department of Pharmaceutical Technology and Biochemistry, Technical University of Gdansk, 90-952 Gdansk, Poland
SOURCE: Journal of Medicinal Chemistry, (1987) 30/10 (1715-1719).
ISSN: 0022-2623 CODEN: JMCMAR
COUNTRY: United States
DOCUMENT TYPE: Journal
FILE SEGMENT: 004 Microbiology
037 Drug Literature Index
LANGUAGE: English

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(FILE 'HOME' ENTERED AT 11:23:12 ON 10 MAR 2005)

FILE 'STNGUIDE' ENTERED AT 11:23:19 ON 10 MAR 2005

FILE 'HOME' ENTERED AT 11:23:26 ON 10 MAR 2005

FILE 'MEDLINE, HCAPLUS, BIOSIS, BIOTECHDS, EMBASE, SCISEARCH' ENTERED AT 11:24:04 ON 10 MAR 2005

L1 1 S N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGUS
L2 1 S N-ACETYGLUCOSAMINE AND CHITIN AND BACTERIA AND FUNGI
L3 708 S CHITIN AND BACTERIA AND FUNGI
L4 531 DUP REM L3 (177 DUPLICATES REMOVED)
L5 69 S L4 AND CHITINOLYTIC
L6 10 S L5 AND (ACETYLHEXOSAMINIDASE OR BETA-1 3 GLUCANASE)
L7 8 S L5 AND ACETYLGLUCOSAMINE
L8 4 S N-ACETYLGLUCOSAMINE FORMATION AND CHITINASE?
L9 4 DUP REM L8 (0 DUPLICATES REMOVED)
L10 69 S N-ACETYLGLUCOSAMINE AND (TRICHODERMA OR APHANOCCLADIUM OR COCC
L11 37 DUP REM L10 (32 DUPLICATES REMOVED)
L12 37 FOCUS L11 1-

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